

House dust mite allergen reduction and allergy at 4 yr: Follow up of the PIAMA-study

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Exposure to high allergen levels in early life is a risk factor for the development of allergy. We previously reported limited effects of mite allergen impermeable mattress covers in the prevention and incidence of asthma and mite allergy (PIAMA) cohort at the age of 1 and 2 yr. We now present the results of follow-up at 4 yr objectives. To examine the effects of early reduction of house dust mite (HDM) allergen exposure by means of mattress covers on the incidence of allergy and asthma symptoms in the PIAMA birth cohort at the age of 4 yr. High-risk children (allergic mother) were prenatally recruited and randomly allocated to three groups; receiving mite allergen impermeable mattress covers ($n = 416$), placebo covers ($n = 394$) or no intervention ($n = 472$). At 4 yr of age, atopy was assessed by questionnaire; specific Immunoglobulin E (IgE) to inhalant and food allergens was measured in serum. Dust samples collected from the children's mattresses were analysed for mite allergens. *Dermatophagoides farinae* 1 allergen (Der f 1) levels in dust were reduced in the active group. However, *Dermatophagoides pteronissinus* 1 (Der p 1) levels, sensitization and atopic symptoms were similar in all groups. We found no effect of mite allergen impermeable mattress covers on sensitization and atopy at 4 yr. Moreover, the allergen reducing effects of the covers had disappeared for one of the two mite allergens that were measured.

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It has been postulated that increased allergen exposure is an important causative factor for the increase in allergic disease in the Western world

(1–5). The use of allergen impermeable mattress covers is potentially effective in reducing house dust mite (HDM) allergen exposure (6). A report of the European community respiratory health survey (ECRHS) has postulated that sensitization to HDM is a risk factor for the development of allergic symptoms such as asthma, atopic dermatitis and rhinitis (7, 8). Various prospective studies have been initiated to investigate to what extent the level of allergen exposure may be responsible for the development of allergic

Abbreviations: HDM: House dust mite ; PIAMA: Prevention and incidence of asthma and mite allergy ; Der p 1: *Dermatophagoides pteronissinus* 1, a dust mite allergen Der f 1: *Dermatophagoides farinae* 1 allergen, a dust mite allergen NHS: Natural history study IS: Intervention study ISAAC: International study of asthma and allergies in children IgE: Immunoglobulin E.

sensitization and atopic phenotypes. In the Manchester Asthma and Allergy Study (MAAS), high-risk children were randomly allocated to a stringent environmental intervention or a normal regime. At the age of 1 yr this intervention resulted in a significant decrease of attacks of severe wheeze with shortness of breath (9). In the Study on the Prevention of Allergy in Europe (SPACE), HDM reduction programs showed an association between HDM allergen exposure and sensitization to mite in the first years of life (10). In the Isle of Wight study, a reduction in current wheeze, night cough, bronchial hyper responsiveness and atopy at the age of 8 yr was found after strict food-allergen avoidance and reduction of HDM exposure in the first 9 months of life (11). We have examined the effects of reduction of mite allergen exposure by mite allergen impermeable mattress covers on the development of sensitization and allergic symptoms in the prevention and incidence of asthma and mite allergy (PIAMA) study. We previously reported that the mattress covers significantly reduced exposure to *Dermatophagoides pteronissinus* 1 (Der p 1) and *Dermatophagoides farinae* 1 allergen (Der f 1) allergen at 1 yr of age, and slightly reduced upper airway symptoms and nocturnal cough without a cold in the first 2 yr of life. However, no evidence of protection against other respiratory symptoms and atopic dermatitis was found. At 1 yr of age, sensitization to mite allergens was similar in the active and the placebo groups (12, 13). We now describe a follow up of the effect of mite impermeable mattress covers on the development of sensitization, wheeze, rhinitis and atopic dermatitis in the PIAMA cohort at 4 yr of age.

Materials and methods

Between March 1996 and May 1997, 10,232 pregnant women were recruited for the PIAMA study. Allergic women, fulfilling the criteria of allergic disease as assessed by a validated questionnaire (14) were invited to participate in a placebo controlled Intervention Study (IS) on the effect of mite allergen impermeable mattress and pillow covers ($n = 855$) or in a natural history study (NHS high-risk) ($n = 472$). In the IS, participants were randomly allocated into two groups: an active group, receiving polyester-cotton mite allergen impermeable mattress and pillow covers (Acb; Allergy Control Products, Saratoga Springs, NY) for the parental and child beds (IS-active, $n = 416$), and a placebo group, receiving cotton placebo covers (IS placebo, $n = 394$) in a double blind fashion. Forty-five partic-

ipants could not be reached or decided to withdraw from the study when the application of the intervention was planned. Children were born between May 1996 and December 1997. The study was approved by the local medical ethical committees and the Dutch Central Committee on Research Involving Human Subjects.

During pregnancy, at the age of 3 months and every year thereafter approximately at the birthday of the child, parents completed questionnaires about allergic symptoms in the child during the previous year. These questionnaires included core questions from the International study of asthma and allergies in childhood (ISAAC) (15). Data on wheezing, cough, rhinitis, atopic dermatitis, medication use, birth characteristics and allergic disease in the parents and siblings were collected by questionnaire. Also, information about general health, indoor environment, socio-economic characteristics, lifestyle, feeding and demographic factors was obtained by questionnaire.

Wheezing, atopic dermatitis, nocturnal dry cough without a cold or chest infection and rhinitis were assessed using the questionnaire data collected in the first 4 yr of life. All children were categorized into four wheezing phenotypes (16): never wheeze, early transient wheeze, persistent wheeze and late wheeze. Rhinitis was defined by questionnaire as a runny nose, not accompanied by a cold, in the past 12 months. We also asked whether the rhinitis was accompanied by eye symptoms.

Total Immunoglobulin E (IgE) and specific IgE against mite, cat, dog and food allergens were determined in serum, obtained by venapuncture with a radio allerge sorbent test (RAST) with a detection limit of 0.15 kU/l as described previously (17). A specific IgE ≥ 0.35 IU/ml was considered positive. Dust samples were collected from the parental and child mattresses and concentrations of mite allergens were measured in a standardized fashion. We previously described the sampling, storage and extraction procedures in detail (18).

Statistical analysis

Data were analysed using the SAS statistical software package v8.2 (Cary, NC, USA). In conformity with the previously presented data of the first 2 yr of life in this study population, the IS-placebo group was compared with the IS-active as well as the NHS high-risk group (12). The level of significance was set at a p -Value of 0.05. General characteristics were analysed by independent sample t -tests or Mann-Whitney

tests depending on the data distribution and, for categorical data, Pearson chi-square tests. Associations between both the IS-placebo/IS-active groups and the IS-placebo/NHS-high risk groups in the development of atopic symptoms were performed by multiple logistic regression analysis, adjusted for gender, mother allergic for HDM, mother ever asthma, maternal smoking during pregnancy, premature birth and pillow encasing used by parents to estimate odds ratio's (OR's) and 95% confidence intervals (CI).

Results

Response

The recruitment and the participation rate during the first 4 yr are summarized in Fig. 1 (12, 13). Participants in the NHS-high risk and the participants in the IS were defined as study population. Participants in the IS receiving no cover were excluded from the analysis. At 4 yr of age, questionnaire data of 1019 of the initial 1282 participating children were obtained (79.5%). Questionnaires were collected from 603 (74.4%) children in the intervention group, receiving

active (324) or placebo (279) covers and from 416 (88.1%) NHS-high risk children.

Children with and without questionnaire data at the age of four were compared. Children still participating at the age of four significantly less frequently had a mother allergic for HDM (44% vs. 53%, $p = 0.03$), a mother who smoked during pregnancy (14% vs. 25%, $p < 0.0001$), a mother (21% vs. 33%, $p = 0.004$), or a father (23% vs. 33%, $p = 0.01$) with a low level of education.

General characteristics of the study population

In Table 1, the general characteristics of the groups are summarized. Although randomization was generally successful, there were a number of differences between the groups. The group with active covers contained fewer boys (46% vs. 58%, $p < 0.05$) and fewer premature

Table 1. General characteristics of the 1019 children with an atopic mother, in the intervention and natural history study

	Active covers (n = 324) %	Placebo covers (n = 279) %	NHS-high risk (n = 416) %
Mother allergy for HDM (by questionnaire)‡	56	57	47‡
Mother ever asthma‡	24	29	22‡
Father allergy (by questionnaire)	35	34	30
Father allergy for HDM (by questionnaire)	15	14	11
Father ever asthma (by questionnaire)	9	10	7
Older sibling allergy	25	21	24
Older sibling ever wheezing	34	34	30
Boys (%)†	46†	58	54
Mother smoked during pregnancy‡	11	12	18‡
Smoke exposure in the home at least 1 day/wk	18	21	23
Born in winter (21 Dec. 1996 to 20 March 1997)	17	20	24
Premature birth (<37 wk of gestation)†	4†	8	5
Breast feeding at 12 wk of age	49	56	51
Other children <12 yr in the household at age 4	86	85	85
Day care at age 4, >4 h per wk	72	72	76
Pets in the house at age 4 yr	48	43	48
Damp- or mould spots in the home	12	10	12
Stop use mattress covers child at 4 yr	13	14	—
Stop use pillow covers child at 4 yr	31	24	—
Stop use mattress covers parents	25	24	—
Mother low level of education	21	21	22
Father low level of education	25	21	23

†Significant difference ($p < 0.05$) between active cover group and placebo group.

‡Significant difference ($p < 0.05$) between natural history group and placebo group.

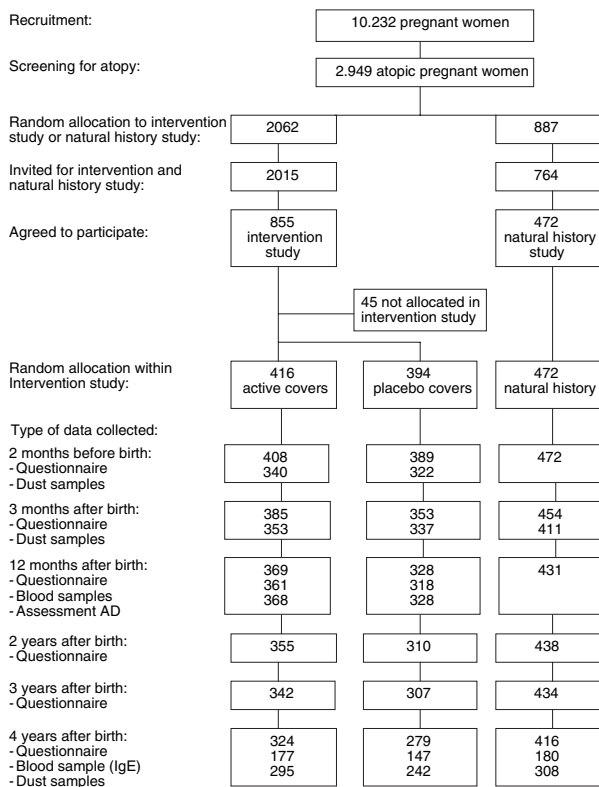


Fig. 1. Prevention and incidence of asthma and mite allergy recruitment flow chart.

births before 37 wk (4% vs. 8%, $p < 0.05$) than the placebo group. In the NHS group, fewer mothers were allergic for HDM (47% vs. 57% $p < 0.05$), fewer mothers reported a diagnosis of asthma (22% vs. 29%, $p < 0.05$) and more mothers had smoked during pregnancy (18% vs. 12%, $p < 0.05$) than in the placebo group.

Compliance to the covers

During the 4-yr follow-up some families stopped using their mattress covers. In Table 2, the compliance of both parents and children is shown. The children's compliance was high for both the active and placebo covers (87% and 86%). Only four children with an active cover and one child with a placebo cover stopped using their cover before the age of two. The compliance of the parents in the active and placebo group was 75% and 76%. Compliance with the use of pillow covers in children was 69% in the active group and 76% in the placebo group. For parents, these were 78% and 85%, respectively ($p < 0.05$).

Twenty-five of the 413 children (6%) participating in the NHS used HDM impermeable mattress covers at the age of 4 yr. In 22 (88%) of these, the covers had been prescribed by a doctor.

Effect of the intervention on house dust mite-allergen exposure

No significant difference in dust exposure was found between the active, the placebo and the natural history group at 4 yr (median of 178, 187

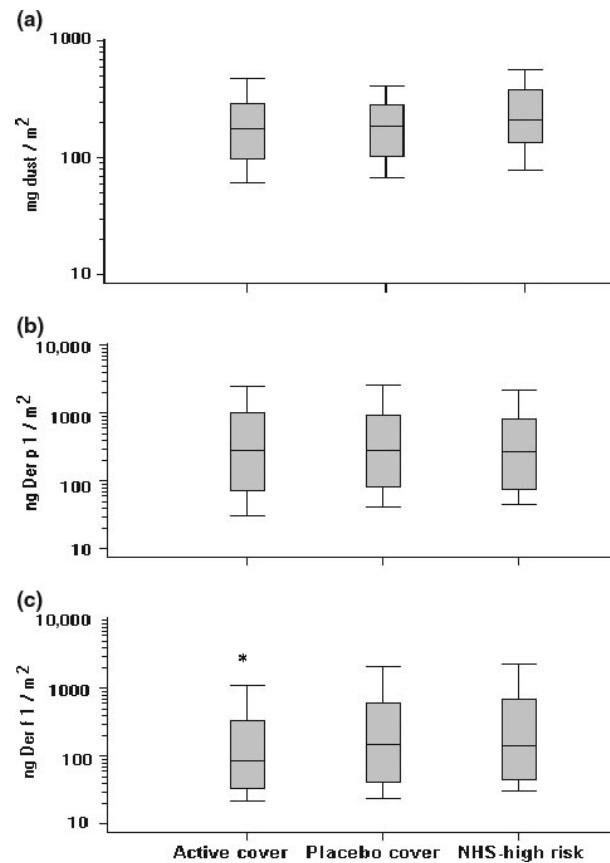


Fig. 2. Box plot of the amount of dust in mg/m^2 (a), the amount of Der p 1 in ng/m^2 (b) and the amount of Der f 1 in ng/m^2 (c) on the children's mattresses in the three groups at 4 yr. Solid lines in box represent median, boxes represent the 25th and 75th percentile and lines represent 10th and 90th percentile. * $p < 0.05$ for difference between active covers and placebo covers.

and $213 \text{ mg dust}/\text{m}^2$, respectively). In Fig. 2a, the box plots of the amounts of dust per m^2 mattress are shown. The median amount of Der p 1 in ng/m^2 was similar in the active, the placebo and the natural history groups (282, 282 and $275 \text{ ng Der p 1}/\text{m}^2$) (Fig. 2b). The level of Der f 1 was significantly lower in the active group [$88 \text{ ng Der f 1}/\text{m}^2$, Interquartile Range (IR) 34–344] as compared with the placebo ($157 \text{ ng Der f 1}/\text{m}^2$, IR 42–625) and the natural history group ($146 \text{ ng Der F 1}/\text{m}^2$, IR 44–719) (Fig. 2c).

Effect of the intervention on the prevalence of clinical symptoms of atopy

In Table 3, the prevalence and adjusted OR's for wheeze, atopic dermatitis and rhinitis in the fourth yr of life and the diagnostic categories of wheeze in the first 4 yr of life are shown. Adjustment was carried out for the following potential confounders: gender, HDM allergy of

Table 2. Compliance to the intervention

	Active covers n = 309 (%)	Placebo covers n = 264 (%)	p-Value
Mattress child			
Stopped at 4 yr (%)	13	14	0.72
Stopped that stopped before 2 yr (%)	10	3	0.22
Pillow child			
Stopped at 4 yr (%)	31	24	0.06
Stopped that stopped before 2 yr (%)	36	23	0.24
Mattress parents			
Stopped at 4 yr (%)	25	24	0.83
Stopped that stopped before 2 yr (%)	29	18	0.17
Pillow parents			
Stopped at 4 yr (%)	22	15	0.04*
Stopped that stopped before 2 yr (%)	38	12	0.009*

Percentage of children and parents who had stopped the use of study covers at the age of 4 yr and percentage of stopped children and parents that stopped before the age of 2 yr.

* $p < 0.05$.

Table 3. Prevalence of atopic symptoms in the different groups, with adjusted odds ratio's and 95% confidence intervals

Atopic phenotype	Prevalence (%)			Comparison of active covers and NHS-high risk vs. placebo covers	
	Placebo covers (n = 279)	Active covers (n = 324)	NHS-High risk (n = 416)	Active covers AOR* (95% CI)	NHS-high risk AOR* (95% CI)
Never wheeze	52	49	56	0.8 (0.5–1.1)	1.1 (0.8–1.5)
Early wheeze	28	27	24	1.0 (0.7–1.5)	0.9 (0.6–1.2)
Late wheeze	3	3	1	1.0 (0.3–2.7)	0.5 (0.1–1.5)
Persistent wheeze	14	13	14	1.3 (0.7–2.1)	1.0 (0.7–1.7)
Wheeze in fourth year	16	17	15	1.2 (0.7–2.0)	0.9 (0.6–1.4)
Cough at night in fourth year	27	27	26	1.0 (0.7–1.5)	1.0 (0.7–1.5)
Atopic dermatitis in fourth year	28	25	20	0.9 (0.6–1.3)	0.7 (0.5–0.9)
Rhinitis in fourth year	28	30	26	1.1 (0.8–1.6)	0.9 (0.6–1.3)
Rhinitis and eye symptoms in fourth year	6	13	10	2.4 (1.2–4.9)	1.5 (0.8–3.0)

Values are presented in percentages for the prevalence of atopic phenotypes and as adjusted odds ratios (with 95% CI) for the relation between group allocation and atopic phenotypes with the placebo group as the index group.

*Logistic regression, adjusted for gender, mother allergic for HDM, mother ever asthma, maternal smoking during pregnancy, premature birth and pillow encasing used by parents.

Table 4. Total and specific IgE at age 4 yr

	Active covers (n = 177)	Placebo covers (n = 147)	NHS-high risk (n = 180)
Total IgE			
Median (IU/ml; interquartile range)	44 (13–101)	48 (16–117)	31 (12–114)
Mean (IU/ml; standard deviation)	132 (386)	115 (219)	149 (305)
Specific IgE to HDM ≥ 0.35 IU/ml/total	28/177 (16%)	22/147 (15%)	30/180 (17%)
Specific IgE to cat ≥ 0.35 IU/ml/total	13/175 (7%)	9/147 (6%)	12/178 (7%)
Specific IgE to dog ≥ 0.35 IU/ml/total	11/174 (6%)	4/146 (3%)	6/178 (3%)
Specific IgE to grasspollen ≥ 0.35 IU/ml/total	14/171 (8%)	6/143 (4%)	16/178 (9%)
Specific IgE to birchpollen ≥ 0.35 IU/ml/total	4/171 (2%)	2/142 (1%)	5/178 (3%)

the mother, history of maternal asthma, maternal smoking during pregnancy, premature birth and pillow covers used by the parents.

Respiratory symptoms. In the active-, the placebo-, and the natural history groups the prevalence of never, early, late and persistent wheeze were not significantly different (Table 3). We also found no significant differences in the prevalence of wheeze, and cough at night without an infection in the fourth year.

Atopic dermatitis. The prevalence and pattern of atopic dermatitis was not significantly different in the active group compared with the placebo group. The prevalence of atopic dermatitis at the age of four was 20% in the natural history group as compared with 28% in the placebo group (AOR 0.7; 95% confidence interval 0.5–0.9).

Rhinitis. The prevalence of a runny nose not accompanied by a cold ever or at the age of four

was not significantly different between the active group, the placebo group and the natural history group. A runny nose with eye symptoms was more frequently reported in the active group than in the placebo group (AOR = 2.6, 95% confidence interval 1.2–5.3).

Effect of the intervention on total and specific IgE

In Table 4, the serum levels of total IgE and specific IgE to HDM, cat dander, dog dander, grass pollen and birch pollen are shown. Median serum levels of total IgE in the three groups were not statistically different. The prevalence of sensitization to inhalant allergens was also similar in the active group, the placebo group and the natural history group. The percentages of children with sensitization were 16%, 15% and 17% for mite and 7%, 6% and 7% for cat, respectively (not significant). In the children sensitized to mite at 4 yr, we found a trend for a lower incidence of

persistent wheeze in the active group compared with the placebo and the natural history group (19%, 33% and 37%, respectively). This was not significant ($p = 0.27$) and numbers were relatively small.

Discussion

Our findings suggest that intervention with mite allergen impermeable mattress covers in the first 4 yr of life did not modify the development of respiratory symptoms, atopic dermatitis, rhinitis and allergic sensitization to an important extent. There was a high compliance with the intervention, but the use of covers no longer reduced exposure to Der p 1 at 4 yr. There was still a modest effect on Der f 1. Sensitization to HDM at 4 yr was similar in children with active and placebo covers.

Several other birth cohort studies have reported effects of allergen reduction measures on the development of atopic phenotypes. In the Isle of Wight study, a reduction in wheeze, night cough and sensitization at the age of 8 yr was found after strict food allergen avoidance and reduction of HDM exposure by the use of an acaricide and mattress covers in the first 9 months of life (11). In the MAAS, a multifaceted intervention to reduce HDM exposure resulted in a substantial reduction of Der p 1 exposure in the first 3.5 yr of life (19). Benefits in terms of reduction of respiratory symptoms were, however, transient and atopic sensitization was not prevented, in fact even higher after the intervention (9, 20). Similarly, the SPACE multi-centre study showed only transient protective effects of mite impermeable mattress covers (21, 22). A Canadian dietary and environmental IS suggested a beneficial effect on asthma symptoms in the first 2 yr of life (23). In the Australian CAPS study, a significant effect of allergen avoidance on the prevalence of sensitization was found, but no differences in the prevalence of wheeze at the age of three (24). We previously reported modest clinical effects of our intervention in the first 2 yr of life, including a reduction in night cough (12). The present data indicate no further beneficial effects on sensitization and respiratory symptoms at 4 yr, suggesting that preventive effects of our intervention were small and transient, whereas no reduction in sensitization was found. The general pattern of most previous studies as well as our present results seems to indicate that single or multifaceted interventions to reduce allergen exposure in early life in high risk children seem to have at best a transient and small effects on atopic symptoms and sensitization. There are

many methodological differences between the studies that may explain apparent discrepancies, especially the nature of the intervention: inhalant allergen reduction with or without food allergen reduction, single- or multi-faceted, placebo-controlled or open. Our study used a double-blind placebo-controlled, simple intervention with mattress and pillow covers. The design was unique in that a placebo arm was included in a blinded fashion, together with a group that got no intervention at all. The randomization of these groups was reasonably successful; there were however slight but significant dysbalances with less allergy or asthma, and more smoking in mothers of the natural history group, and there were fewer boys and pre-matures in the active intervention group that could potentially have affected the results. We would expect no differences in the prevalence of sensitization and atopic symptoms between the placebo and the NHS-high risk group, but surprisingly the prevalence of atopic dermatitis in the NHS-high risk group was lower than in the placebo group. This could be explained by a lower percentage of HDM allergic mothers in the NHS-high risk group as a result of the suboptimal randomization, which may have resulted in a group genetically less prone to develop atopy and sensitization to HDM, but this is not confirmed by the other diagnostic criteria of atopy in this study. Although the prevalence of rhinitis was similar in the groups, in the active cover group the children diagnosed with rhinitis had significantly more often a combination with eye symptoms. This could be a result of the intervention. The clinical diagnosis of asthma is uncertain in the first year of life. For this reason, we categorized different wheezing phenotypes. There was, however, no phenotype-specific benefit of our intervention. The cohort will be followed until the age of 8 yr, at that age it will be possible to make a reliable diagnosis of asthma and to know whether the intervention by HDM allergen impermeable mattress covers has had any protective effects on the long term.

Despite our negative results, in our opinion the application of covers has not been invalidated yet. The HDM levels in the first months of the PIAMA cohort were low as compared with those in other intervention studies, and as compared with our previous studies among infants in The Netherlands (25–27). We could attribute this unfortunate fact to highly unusual climatic conditions in the years of recruitment (28). This also explains why exposure to HDM was 4–5-fold higher at 4 yr compared with 3 months (26). With these initially low mite allergen levels, our

intervention could only have a limited impact on allergen exposure.

In conclusion, after 4 yr of use, HDM allergen impermeable mattress covers had no effect on the exposure to HDM and Der p 1, while Der f 1 levels remained reduced. We found no effect of the mattress covers on wheeze and other respiratory symptoms and sensitization at age 4 yr between the intervention-, placebo- and the natural history groups. Hence, we conclude that in this cohort allergen impermeable mattress covers applied before birth were not effective in preventing sensitization and respiratory symptoms in high-risk children at the age of 4 yr.

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