Cooper, Philip J; Chico, Martha E; Amorim, Leila D; Sandoval, Carlos; Vaca, Maritza; Strina, Agostino; Campos, Ana Clara; Rodrigues, Laura C; Barreto, Mauricio L; Strachan, David P; (2015) Effects of maternal geohelminth infections on allergy in early childhood. The Journal of allergy and clinical immunology, 137 (3). 899-906.e2. ISSN 0091-6749 DOI: https://doi.org/10.1016/j.jaci.2015.07.044

Downloaded from: http://researchonline.lshtm.ac.uk/id/eprint/2312481/

DOI: https://doi.org/10.1016/j.jaci.2015.07.044

Usage Guidelines:

Please refer to usage guidelines at https://researchonline.lshtm.ac.uk/policies.html or alternatively contact researchonline@lshtm.ac.uk.

Available under license: http://creativecommons.org/licenses/by/2.5/
Effects of maternal geohelminth infections on allergy in early childhood

Philip J. Cooper, MB BS, PhD, a,b,c Martha E. Chico, MD, a Leila D. Amorim, PhD, d Carlos Sandoval, TMD, d
Maritza Vaca, MD, a Agostino Strina, MD, a Ana Clara Campos, MSc, a Laura C. Rodrigues, MD, PhD, f
Mauricio L. Barreto, MD, PhD, a and David P. Strachan, MD, FRCP d
Quininde and Quito, Ecuador, London, United Kingdom, and Salvador, Brazil

Background: Maternal geohelminth infections during pregnancy may protect against allergy development in childhood.

Objective: We sought to investigate the effect of maternal geohelminths on the development of eczema, wheeze, and atopy during the first 3 years of life.

Methods: A cohort of 2404 neonates was followed to 3 years of age in a rural district in coastal Ecuador. Data on wheeze and eczema were collected by means of questionnaire and physical examination at 13, 24, and 36 months of age. Atopy was measured based on skin prick test (SPT) reactivity to 9 allergens at 36 months. Maternal stool samples were examined for geohelminths by microscopy. Data on potential confounders was collected after birth by questionnaire.

Results: Geohelminths were observed in 45.9% of mothers. Eczema and wheeze were reported for 17.7% and 25.9%, respectively, of 2069 (86.1%) children with complete follow-up to 3 years, and allergen SPT reactivity to any allergen was present in 17.2% and to house dust mite in 8.7%. Maternal geohelminth infections were not significantly associated with eczema (adjusted odds ratio [OR], 1.26; 95% CI, 0.98-1.61), wheeze (adjusted OR, 1.02; 95% CI, 0.82-1.27), and SPT reactivity to any allergen (adjusted OR, 0.79; 95% CI, 0.61-1.01). In subgroup analyses maternal geohelminths were associated with a significantly reduced risk of SPT reactivity to mite and other perennial allergens, and maternal ascariasis was associated with an increased risk of eczema and reduced risk of SPT reactivity to all allergens.

Conclusion: Our data do not support a protective effect of maternal infections with geohelminth parasites during pregnancy against the development of eczema and wheeze in early childhood, although there was evidence in subgroup analyses for a reduction in SPT reactivity to house dust mites and perennial allergens. (J Allergy Clin Immunol 2016;137:899-906.)

Key words: Geohelminths, maternal infections, atopy, wheeze, eczema, early childhood

Allergic diseases, such as asthma and eczema, have emerged as major health problems in many developing countries, particularly in urban Latin America. The causes of such temporal increases are likely to be explained by environmental changes.

The hygiene hypothesis has explained the increasing prevalence of allergic diseases in the context of decreasing exposures to common infectious diseases of childhood and a decreased diversity of microbial exposures in the environment. Among potentially protective infections, there has been considerable interest in the role of geohelminth parasites (e.g., Ascaris lumbricoides, Trichuris trichiura, and hookworm), which are extremely common chronic infections of childhood in developing countries, where an estimated 2 billion humans are infected. The relationship between geohelminth infections and allergy is complex. Although infections are generally associated with a reduced prevalence of allergen skin prick test (SPT) reactivity, their effects on allergic diseases are less clear. Treatment studies of geohelminths in schoolchildren have not shown effects on the prevalence of allergic diseases or consistent increases in allergen SPT reactivity after anthelmintic treatment. It has been suggested that in utero and early-life exposures might be important for mediating protection. Other environmental exposures that have been associated with protection against allergy, such as farming, have stronger effects when exposure occurs in utero and in early childhood.

We recruited a birth cohort in an area of Ecuador with a high maternal prevalence of geohelminth infections to test the hypothesis that in utero exposures to geohelminth parasites through having an infected mother protect against the development of eczema, wheeze, and atopy during the first 3 years of life.

METHODS

Study design and setting

A prospective study from birth was done in the rural District of Quininde, Esmeraldas Province, Ecuador, as described in detail elsewhere. The district serves a population of approximately 150,000 with an economy based on agricultural activities, primarily African palm oil, and limited access to basic services.
Study participants

Neonates were recruited in the public hospital, Hospital “Padre Alberto Bumf,” which serves the district, between November 2005 and December 2009. Eligibility criteria were as follows: (1) healthy baby aged less than 14 days; (2) collection of stool sample from the mother; (3) mother aged 17 years or more; (4) family resident in the District of Quininde for at least 2 years with plans to stay for at least 3 more years; and (5) accessible household.

Questionnaires

Children were followed up to 3 years of age. Data on demographic and socioeconomic factors, household characteristics, maternal obstetric history, family history of chronic illnesses, and relevant environmental exposures within and outside the household were collected using an interview-led questionnaire from the child’s mother after the child’s birth. Questionnaires were done annually at 13, 24, and 36 months to collect data on the development of eczema and wheezing and other relevant information.

Measurement of maternal infections with geohelminth infections

Stool samples to measure geohelminth infections were collected from mothers during the third trimester or immediately after the child’s birth and in children at 13 and 24 months of age. Samples were examined using a combination of methods, including saline mounts, the Kato-Katz method, formol-ether concentration, and carbon coproculture methods. A positive sample was defined by the presence of at least 1 egg or larva from any of the 4 detection methods. A *Lumbricoides* and *Trichuriasis* infection intensities were expressed as eggs per gram (epg) of feces using results from the Kato-Katz method.

Measurement of eczema and wheeze

Data were collected using periodic questionnaires and examinations for visible signs of flexural dermatitis with a standardized photographic protocol. Eczema was defined using the United Kingdom refinement of the Hanifin and Rajka diagnostic criteria. Children were considered to have eczema if the mother reported the child to have had an itchy skin condition during the previous 12 months plus at least 3 of the following: (1) involvement of skin creases and cheeks; (2) history of allergic disease in siblings or parents; (3) history of generally dry skin; and (4) visible flexural dermatitis, including that affecting the cheeks, forehead, and outer limbs. Recent wheeze was defined as any episode of reported wheeze during the previous 12 months.

Allergen SPT reactivity

Allergic sensitization was measured at 36 months using SPTs with 9 allergen extracts (Greer Laboratories, Lenoir, NC): house dust mite (HDMs; *Dermatophagoides pteronyssinus*/Dermatophagoides farinae mix), American cockroach (*Periplaneta americana*), cat, dog, grass pollen (9 southern grass mix), fungi (new stock fungi mix), egg, milk, and peanut, with positive histamine and negative saline controls. A positive reaction was defined as a mean wheal diameter of at least 2 mm greater than that elicited by the saline control 15 minutes after pricking the allergen onto the volar side of the forearm with ALK-Abelló lancets (ALK-Abelló, Hungerford, United Kingdom). A positive SPT response was defined as a positive reaction to any of the allergens tested. All testing was done by trained physicians (M.E.C. and M.V.).

Statistical analysis

We estimated that with 1840 children followed up at 3 years of age and approximately 50% of mothers infected with geohelminths, we would have greater than 95% power to detect a difference of at least 5% between groups for outcomes with 10% or greater prevalence, considering a P value of less than .05. The primary analysis was the association between maternal geohelminths and eczema, wheeze, and atopy. Secondary analyses addressed associations between maternal geohelminth species and study outcomes, including atopy atopy. Bivariate logistic regression analyses were done to estimate associations between maternal geohelminth infections or potential confounders and outcomes. Multivariate logistic regression was used to estimate associations between maternal geohelminth infections and outcomes. Potential confounders considered in the analyses are shown in Table I. A socioeconomic status index was created by using principal components analysis of 7 socioeconomic variables, as previously described. Child geohelminth infections were defined as any infection during the first 2 years of life. Potential confounders in any of the bivariate analyses with a P value of less than .20 were kept in the final models using the same set of confounders to adjust all models. All statistical analyses were done with Stata 11 software (StataCorp, College Station, Tex).

Ethical considerations

The protocol was approved by the ethics committees of the Hospital Pedro Vicente Maldonado, Universidad San Francisco de Quito, and Pontificia Universidad Catolica del Ecuador. The study is registered as an observational study (ISRCTN41239086). Informed written consent was obtained from the child’s mother. Anthelmintic treatment (single dose of 400 mg of albendazole) was provided to mothers with geohelminth infections after delivery. Children with positive stool results for geohelminths were treated with a single dose of 400 mg of albendazole if aged 2 years or greater and with pyrantel pamoate (11 mg/kg) if aged less than 2 years, according to Ecuadorian Ministry of Public Health recommendations.

RESULTS

Cohort participants

Follow-up information for the 2404 neonates recruited is provided in Fig 1. Follow-up was greater than 90% at each of the annual follow-ups during the first 3 years of life in the cohort, and complete data for all 3 observation times (ie, 13, 24, and 36 months) were available for 2069 (86.1%) children. Maternal infections with malaria, HIV, and other helminths were of low prevalence (<0.5%) in the study population.

Frequencies of exposures and outcomes

Geohelminth infections were detected in 45.9% of mothers as follows: *Lumbricoides* (27.5%), *Trichuriasis* (28.9%), hookworm (5.6%), and Strongyloides stercoralis (3.9%). The only other helminth parasite observed was the cestode *Hymanolepis nana* (0.5%). The proportions of mothers by infection intensity category were as follows: *Lumbricoides* light (<5000 epg) at 17.1% and moderate/heavy (>5000 epg) at 5.1% and *Trichuriasis* light (<1000 epg) at 22.0% and moderate/heavy (>1000 epg) at 4.2%. The prevalence of eczema at 13, 24, and 36 months was 12.6%, 4.7%, and 3.4%, respectively, with 17.7% of children having eczema at least once. Only 2.5% had episodes of eczema documented for 2 or more visits. The prevalence of reported wheeze at 13, 24, and 36 months was 16.1%, 8.0%, and 8.5%, respectively, and 25.9% had at least 1 reported episode. Allergen SPT reactivity was present in 17.2% of children at 3 years of age: *D. pteronyssinus*/farinae (8.7%), cockroach (2.9%), mixed...
fungi (2.4%), dog (2.3%), cat (1.2%), mixed grasses (2.5%), peanut (1.2%), milk (1.2%), and egg (1.9%).

Determinants of wheeze, eczema, and allergen SPT reactivity between birth and 3 years of age

The distributions of demographic and potential confounding variables are shown in Table I. Most mothers were of Mestizo ethnicity (73.4%), had at least completed primary education (73.4%), and lived in a neighborhood (70.5%) of one of the towns (denoted urban) in the district. Almost a third (28.9%) of children acquired at least 1 geohelminth infection during the first 2 years of life: *A lumbricoides* in 22.6% and *T trichiura* in 12.7%. Prevalence of any geohelminth infections at 13 and 24 months were 12.3% and 21.8%, respectively. Table I shows results of bivariate analyses between exposures or potential confounders.
and study outcomes. Results of multivariate analyses are shown in Table II. None of the outcomes in children up to or at 3 years of age were statistically significantly associated with maternal geohelminth infections after adjustment for potential confounders. With respect to other risk factors, maternal allergy (odds ratio [OR], 2.29), household pets (OR, 1.32), and infantile pneumonia (OR, 2.05) were associated with eczema. Having an Afro-Ecuadorian mother (vs others; OR, 1.30) and infantile pneumonia (OR, 5.64) were associated with wheeze. SPT response positivity to any allergen was not significantly associated with any of the variables analyzed. Infection with any geohelminth parasite during the first 2 years of life was not associated with any of the outcomes measured at 3 years of age.

For the analysis presented here, we considered only complete data for all observation times to reduce classification bias for those with incomplete data. We compared risk factors between the subjects included in the analysis with those with incomplete information and found no substantial differences between the 2 groups. Thus we can assume that missing data were random and should not have led to bias.

**Effects of maternal geohelminths on SPT reactivity to HDMs and perennial allergens**

Because HDMs are the dominant allergens in our study population, we analyzed separately the associations of maternal geohelminths and potential confounders with SPT reactivity to HDM and all perennial allergens tested. Results of crude and adjusted analyses are provided in Table E1 in this article’s Online Repository at www.jacionline.org. In adjusted analyses maternal geohelminth infections were significantly inversely associated with SPT reactivity to HDM (OR, 0.61) and all perennial allergens (OR, 0.69). SPT reactivity to HDM was independently associated with maternal contact with large farm animals during pregnancy (OR, 0.72).

**Effects of individual geohelminth parasites on study outcomes**

Table E2 in this article’s Online Repository at www.jacionline.org shows the results of crude and Table III shows the results of adjusted analyses of the associations between individual geohelminth species detected in the mother and outcomes. In adjusted analyses *A lumbricoides* infection was significantly associated with an increased prevalence of early childhood eczema (OR, 1.36) but a lower prevalence of SPT reactivity to any allergen (OR, 0.70) and HDM (OR, 0.48). There was a borderline significantly increased risk of wheeze associated with *A lumbricoides* infection (OR, 1.27). *T trichiura* and hookworm infections were not associated with eczema, wheeze, or SPT response positivity to any allergen or to HDM. There was no evidence of stronger effects at higher parasite burdens of maternal *A lumbricoides*. 

**FIG 1.** Participant flow through follow-up to 3 years of age and those included in the analysis.
**DISCUSSION**

In this prospective study done from birth to 3 years of age in a rural district of Esmeraldas Province in a tropical region of Ecuador, we have examined the hypothesis that maternal geohelminth infections during pregnancy protect against the development of atopy (measured using SPTs), eczema, and wheeze in early childhood. Overall, our data indicate that maternal geohelminths do not protect against eczema and wheeze in early childhood. In subgroup analyses there was evidence that maternal geohelminths protected against SPT reactivity to mite and other perennial allergens and that maternal ascariasis increased the risk of eczema but reduced the prevalence of allergen SPT reactivity. Helminth parasites are potent modulators of the host immune response. Their modulatory effects are directed largely against allergic inflammatory mechanisms that serve to kill the parasites and prevent establishment of chronic infections or control parasite numbers once chronic infections have become established. It is widely believed that the immune modulation associated with chronic helminth infections might also protect against allergic inflammation and allergic diseases. Numerous studies have shown geohelminths to be inversely associated with SPT response positivity in schoolchildren, although the effects of geohelminths on allergic diseases are less consistent. The treatment of geohelminths does not appear

### TABLE II. Adjusted analyses for associations between maternal geohelminth infections or potential confounders and study outcomes

| Variable | Eczema to 3 y | Wheeze to 3 y | Allergen SPT reactivity at 3 y |
|----------|--------------|--------------|--------------------------------|
|          | Adjusted OR (95% CI) | P value | Adjusted OR (95% CI) | P value | Adjusted OR (95% CI) | P value |
| Any maternal geohelminth | | | | | | |
| No | 1 | | | | | |
| Yes | 1.26 (0.98-1.61) | .067 | 1.02 (0.82-1.27) | .837 | 0.79 (0.61-1.01) | .060 |
| Maternal ethnicity | | | | | | |
| Non–Afro-Ecuadorian | 1 | | | | | |
| Afro-Ecuadorian | 1.25 (0.96-1.62) | .103 | 1.30 (1.03-1.65) | .026 | 1.12 (0.85-1.47) | .413 |
| Maternal educational level | | | | | | |
| Illiterate | 1 | | | | | |
| Complete primary | 0.83 (0.58-1.16) | .275 | 0.87 (0.64-1.18) | .368 | 0.80 (0.56-1.14) | .225 |
| Complete secondary | 0.98 (0.64-1.49) | .915 | 0.89 (0.61-1.30) | .540 | 0.67 (0.43-1.04) | .073 |
| Area of residence | | | | | | |
| Urban | 1 | | | | | |
| Rural | 1.07 (0.81-1.43) | .627 | 0.91 (0.71-1.18) | .477 | 1.00 (0.75-1.34) | .987 |
| Sex | | | | | | |
| Male | 1 | | | | | |
| Female | 0.90 (0.71-1.14) | .373 | 0.77 (0.63-0.95) | .014 | 0.91 (0.72-1.15) | .439 |
| Socioeconomic status* | | | | | | |
| 1 | 1 | | | | | |
| 2 | 0.86 (0.64-1.15) | .301 | 0.84 (0.65-1.09) | .188 | 1.03 (0.77-1.39) | .827 |
| 3 | 0.85 (0.62-1.17) | .313 | 1.04 (0.79-1.37) | .785 | 1.05 (0.76-1.46) | .750 |
| Birth order | | | | | | |
| First | 1 | | | | | |
| Second-fourth | 1.09 (0.81-1.46) | .573 | 1.26 (0.97-1.64) | .080 | 0.87 (0.64-1.14) | .287 |
| Fifth | 0.85 (0.57-1.28) | .441 | 1.18 (0.83-1.67) | .357 | 0.74 (0.50-1.10) | .137 |
| Maternal allergy | | | | | | |
| No | 1 | | | | | |
| Yes | 2.29 (1.46-3.61) | .001 | 1.34 (0.85-2.12) | .208 | 1.33 (0.80-2.23) | .276 |
| Household overcrowding† | | | | | | |
| No | 1 | | | | | |
| Yes | 1.25 (0.96-1.61) | .094 | 1.15 (0.91-1.44) | .239 | 1.08 (0.83-1.14) | .564 |
| Pets inside house | | | | | | |
| No | 1 | | | | | |
| Yes | 1.32 (1.02-1.71) | .036 | 1.15 (0.92-1.46) | .221 | 0.98 (0.75-1.29) | .909 |
| Contact with large farm animals‡ | | | | | | |
| No | 1 | | | | | |
| Yes | 0.94 (0.72-1.22) | .653 | 0.88 (0.70-1.11) | .291 | 0.79 (0.60-1.04) | .089 |
| Pneumonia to 13 mo | | | | | | |
| No | 1 | | | | | |
| Yes | 2.05 (1.27-3.29) | .003 | 5.64 (3.61-8.81) | <.001 | 0.59 (0.30-1.15) | .122 |
| Geohelminth infections in child§ | | | | | | |
| No | 1 | | | | | |
| Yes | 0.98 (0.75-1.28) | .869 | 1.22 (0.97-1.54) | .096 | 0.92 (0.70-1.21) | .542 |

ORs and 95% CIs were estimated using logistic regression and adjusted for all variables shown. P values of less than .05 are shown in boldface. SPT, Allergen skin prick test reactivity to any of 10 allergens.

* Socioeconomic status (SES) represents tertiles of z scores obtained by using a factor analysis, with 1 representing the lowest and 3 representing the highest SES.

† Household overcrowding is defined as the number of persons living in the household per sleeping room.

‡ Any of cows, pigs, mules, donkeys, and horses.

§ Geohelminth infections in the child: presence of any geohelminth infection detected during the first 2 years of life.
to affect the prevalence of allergic diseases but has been shown to increase allergen SPT reactivity in some but not other studies. One explanation for the inconsistent findings of randomized treatment studies on allergen SPT reactivity is the presence of different geohelminth parasites in different populations that might have distinct effects on allergy. Another is that the different geohelminth parasites in different populations that might have distinct effects on allergy. Another is that the different geohelminth parasites in different populations that might have distinct effects on allergy. Another is that the different geohelminth parasites in different populations that might have distinct effects on allergy.

Possible explanations for differences in findings between the Ugandan study and our own include (1) effects of maternal albendazole on organisms other than hookworm or a direct effect on the developing immune system and (2) helminth-specific effects: hookworm, which infected only 5.6% of mothers in our study, and was the predominant geohelminth in the Ugandan study (44% of mothers), appears to protect against the risk of asthma in older children and might also reduce the risk of eczema in infancy and might be the only common geohelminth with protective effects against allergic diseases.

We have shown previously in a cross-sectional analysis of schoolchildren in the same Ecuadorian province that childhood infections with geohelminths did not protect against symptoms of eczema, rhinitis, and wheeze but were inversely associated with allergen SPT reactivity. However, the risk of eczema was associated with poor hygiene exposures, whereas a study in urban Brazil showed that childhood asthma was associated with living in “dirty” households. Symptoms of wheeze and rhinitis were associated with poor hygiene exposures, whereas a study in urban Brazil showed that childhood asthma was associated with living in “dirty” households. Symptoms of so-called allergic diseases in these populations were weakly associated with allergen SPT reactivity. Such attenuation might be explained by the immune-modulatory effects of early-life exposures to geohelminths and a microbially diverse environment.

In analyses of subgroups within the cohort, we observed a protective effect for maternal geohelminths against the development of SPT reactivity to HDM and other perennial allergens and for maternal ascariasis against SPT reactivity to any allergen at 3 years of age. These effects were not related to infection

### TABLE III. Adjusted analyses for associations between types of maternal geohelminth infections and study outcomes, including SPT reactivity to HDMs

| Geohelminth variable | Eczema to 3 y | Wheeze to 3 y | Allergen SPT reactivity at 3 y | SPT reactivity to HDM at 3 y |
|----------------------|--------------|--------------|-------------------------------|-----------------------------|
|                      | Percent      | OR (95% CI)  | Percent                       | OR (95% CI)                  | Percent                       | OR (95% CI)                  | Percent                       | OR (95% CI)                  |
| Maternal *A. lumbricoides* infection |               |              |                               |                             |                             |                             |
| Presence              |              |              |                               |                             |                             |                             |
| No (72.5%)            | 16.2         | 1            |                               |                             |                             |                             |
| Yes (27.5%)           | 21.8         | **1.36 (1.04-1.77)** | **.026**                      | **18.3**                    | **1.70 (0.52-0.95)** | **.021**                      |
| Intensity             |              |              |                               |                             |                             |                             |
| Uninfected (77.8%)    | 16.6         | 1            |                               |                             |                             |                             |
| Light (17.1%)         | 21.7         | 1.32 (0.97-1.81) | .075                         | 18.0                        | 1                           |                             |
| Moderate/heavy (51.1%)| 21.0         | 1.23 (0.71-2.10) | .459                         | 16.2                        | 0.97 (0.54-1.73) | .918                         |
| Maternal *T. trichiura* infection |              |              |                               |                             |                             |                             |
| Presence              |              |              |                               |                             |                             |                             |
| No (71.1%)            | 16.9         | 1            |                               |                             |                             |                             |
| Yes (28.9%)           | 17.7         | 1.11 (0.84-1.45) | .463                         | 17.8                        | 1                           |                             |
| Intensity             |              |              |                               |                             |                             |                             |
| Uninfected (73.8%)    | 17.0         | 1            |                               |                             |                             |                             |
| Light (22.0%)         | 18.9         | 1.04 (0.77-1.39) | .811                         | 17.8                        | 1                           |                             |
| Moderate/heavy (4.2%) | 24.7         | 1.48 (0.84-2.60) | .167                         | 12.9                        | 0.81 (0.41-1.61) | .554                         |
| Maternal hookworm presence |              |              |                               |                             |                             |                             |
| No (94.4%)            | 17.4         | 1            |                               |                             |                             |                             |
| Yes (5.6%)            | 24.4         | 1.35 (0.85-2.16) | .205                         | 23.5                        | 0.71 (0.44-1.14) | .153                         |

*ORs and 95% CIs were estimated using logistic regression. $P$ values of less than .05 are shown in boldface. ORs are controlled for maternal ethnicity and educational level, area of residence, sex, socioeconomic status, birth order, maternal allergy, household overcrowding, pets inside the house, contact with large farm animals, pneumonia to 13 months, and the presence of other types of maternal geohelminth parasites. Uninfected data for prevalence are derived from the results of all parasitologic methods, whereas those for infection intensity were derived from Kato-Katz results.*
The programme of research. SCAALA (Social Changes, Asthma and Allergies in Latin America) is a longitudinal study of asthma and allergies in childhood.30 Previous studies that have shown that living on a farm during pregnancy protects against atopy and allergic diseases,13,14 an effect attributed to the wide variety of microbial exposures of farm residence,29 leading perhaps to the induction of robust immune regulation.4

The strengths of the study include its prospective design and the collection of a large number of sociodemographic and lifestyle variables allowing us to control for potential confounders: the fact that ORs were affected little by adjustment indicates minimal residual confounding. Potential biases were reduced by using objective measures of geohelminth infection, performing all evaluations blind to the child’s exposure status, and high rates of retention in the cohort to 3 years of age. Repeated collection of data over the 3 years by experienced and trained clinicians provided more precise estimates of the prevalence of wheeze and eczema. Some degree of misclassification of eczema with other skin conditions, such as scabies, cannot be excluded, although the eczema definition used here18 might be more specific than other definitions, such as that used by the International Study of Asthma and Allergies in Childhood.30 Previous studies that have addressed the effects of geohelminths during early childhood on the development of eczema or wheeze in early childhood either were unable to measure associations31 or had limited power24 because of low prevalence. Maternal geohelminth infections were measured in the third trimester or in a stool sample collected around the time of birth, the latter being a reasonable proxy for the former because these infections are chronic and can survive in the human intestinal tract for several years.

In summary, we have examined the effects of exposures to geohelminths in pregnancy and early childhood on the development of eczema, wheeze, and allergen SPT reactivity. Our data do not support the hypothesis that early-life exposures to geohelminths protect against the development of eczema and wheeze. Similarly, there was no significant overall protective effect of maternal geohelminths against allergen SPT reactivity, although in subgroup analyses there was evidence for protection against SPT reactivity to HDM and perennial allergens and that maternal ascariasis protected against allergen SPT reactivity, the clinical significance of which remains to be defined and will require replication in future studies.

We thank the ECUAVIDA study team for their dedicated work and the cohort mothers and children for their enthusiastic participation. We acknowledge also the support of the Directors and staff of the Hospital “Padre Alberto Buffoni” in Quinindé, Esmeraldas Province. The study forms part of the SCAALA (Social Changes, Asthma and Allergies in Latin America) programme of research.

Key messages
• In utero exposures to geohelminths are considered to protect against allergy and contribute to the low prevalence of allergic diseases in the rural tropics.
• Data from a birth cohort in Ecuador did not show a significant protective effect of maternal geohelminths against the development of eczema, wheeze, and atopy during the first 3 years of life.
• Subgroup analyses indicated a possible protective effect of maternal geohelminths against atopy to perennial allergens, particularly mite.

REFERENCES
1. Cooper PJ, Rodrigues LC, Cruz AA, Barreto M. The asthma epidemic in Latin America: a public health challenge and research opportunities. Allergy 2009;64:5-17.
2. Eder W, Ege MJ, von Mutius E. The asthma epidemic. N Engl J Med 2006;355:2226-35.
3. Strachan DP. Hay fever, hygiene, and household size. BMJ 1989;299:1259-60.
4. Rook GA. Regulation of the immune system by biodiversity from the natural environment: an ecosystem service essential to health. Proc Natl Acad Sci U S A 2013;110:18360-7.
5. Wammes LJ, Mpairwe H, Elliott AM, Yazdanbakhsh M. Helminth therapy or elimination: epidemiological, immunological, and clinical considerations. Lancet Infect Dis 2014;14:1150-62.
6. Feary J, Britton J, Leonard-See J. Atopy and current intestinal parasite infection: a systematic review and meta-analysis. Allergy 2011;66:569-78.
7. Leonard-See J, Pritchard D, Britton J. The Parasites in Asthma Collaboration. Asthma and current intestinal parasite infection: a systematic review of comparative epidemiological studies. Am J Respir Crit Care Med 2006;174:514-23.
8. Cooper PJ, Chico ME, Vaca MG, Moncayo AL, Bland JM, Mafla E, et al. Impact of a monthly treatment of geohelminth-infected children with albendazole on atopy prevalence: a cluster-randomized trial. Lancet 2006;367:5983-96.
9. Floro C, Tuyen LN, Quinnell RJ, Lewis S, Minh HT, Campbell J, et al. Reduced helminth burden increases allergen skin sensitization but not clinical allergy: a randomized, double-blind, placebo-controlled trial in Vietnam. Clin Exp Allergy 2010;40:111-42.
10. Wirta AE, Hamid F, Wammes LJ, Kaisar MM, May L, Prasetyani MA, et al. The effect of three-monthly albendazole treatment on malarial parasitism and allergy: a household-based cluster-randomized, double-blind, placebo-controlled trial. PLoS One 2013;8:e57899.
11. van den Biggelaar AH, Rodrigues LC, van Ree R, van der Zee JS, Hoeksma-Kruize VY, Souverijn HJ, et al. Long-term treatment of intestinal helminths increases mite skin-test reactivity in Gabonese schoolchildren. J Infect Dis 2004;189:892-900.
12. Elliott AM, Mpairwe H, Quigley MA, Nampijja M, Muhangi L, Oweka-Onyee J, et al. Helminth infection during pregnancy and development of infantile eczema. JAMA 2005;294:2032-4.
13. Douwes J, Cheng S, Travier N, Cohen C, Niesink A, McKenzie J, et al. Farm exposure in utero may protect against asthma, hay fever and eczema. Eur Respir J 2008;32:603-11.
14. von Mutius E. 99th Dahlem conference on infection, inflammation and chronic inflammatory disorders: farm lifestyles and the hygiene hypothesis. Clin Exp Immunol 2010;160:130-5.
15. Wasiuki G, Vercelli D. The farm effect, or: when, what and how a farming environment protects from asthma and allergic disease. Curr Opin Allergy Clin Immunol 2012;12:461-6.
16. Cooper PJ, Chico ME, Platts-Mills TA, Rodrigues LC, Strachan DP, Barreto ML. Cohort Profile: the Ecuador Life (ECUA VIDA) study in Esmeraldas Province, Ecuador. Int J Epidemiol 2014 Jul 2. pii: dyu128. [Epub ahead of print].
17. World Health Organization. Diagnostic techniques for intestinal parasitic infections (IP) applicable to primary health care (PHC) services. Geneva: World Health Organization; 1985.
18. Williams HC, Floro C. So how do I define atopic eczema? Available at: http://www.nottingham.ac.uk/dermatology/eczema/. Accessed May 27, 2014.
19. Menzies SK, Rodriguez A, Chico M, Sandoval C, Broncano N, Guadalupe I, et al. Risk factors for soil-transmitted helminth infections during the first 3 years of life in the tropics; findings from a birth cohort. PLoS Negl Trop Dis 2014;8: e2718.

20. Calvopiña M. Terapéutica antiparasitaria. 2nd ed. Quito (Ecuador): Ministerio de Salud Pública del Ecuador; 1997.

21. Ministerio de Salud Pública del Ecuador. Atencion integrada a las enfermedades prevalentes de la Infancia: cuadros de procedimientos. Quito (Ecuador): Dirección de Promoción y Atención Integral de Salud; 2011.

22. McSorley HJ, Hewitson JP, Maizels RM. Immunomodulation by helminth parasites: defining mechanisms and mediators. Int J Parasitol 2013;43:301-10.

23. Cooper PJ. Interactions of parasites with allergy. Curr Opin Allergy Clin Immunol 2009;9:29-37.

24. Ndibazza J, Mpairwe H, Webb EL, Mawa PA, Nampijja M, Muhangi L, et al. Impact of anthelminthic treatment in pregnancy and childhood on immunisations, infections and eczema in childhood: a randomised controlled trial. PLoS One 2012;7:e50325.

25. Mpairwe H, Webb EL, Muhangi L, Ndibazza J, Akishule D, Nampijja M, et al. Anthelminthic treatment during pregnancy is associated with increased risk of infantile eczema: randomised-controlled trial results. Pediatr Allergy Immunol 2011;22:305-12.

26. Cooper PJ, Vaca M, Rodriguez A, Chico ME, Santos DN, Rodrigues LC, et al. Hygiene, atopy and wheeze-eczema-rhinitis symptoms in schoolchildren from urban and rural Ecuador. Thorax 2014;69:232-9.

27. Barreto ML, Cunha SS, Fiaccone R, Esquivel R, Amorim LD, Alvim S, et al. Poverty, dirt, infections and non-atopic wheezing in children from a Brazilian urban center. Respir Res 2010;11:67.

28. Rodrigues LC, Newcombe PJ, Cunha SS, Alcantara-Neves NM, Genser B, Cruz AA, et al. Early infections with intestinal helminths reduce the risk of atopy later in childhood. Clin Exp Allergy 2008;38:1769-77.

29. Ege MJ, Mayer M, Normand AC, Genuneit J, Cookson WO, Braun-Fahrlander C, et al. Exposure to environmental microorganisms and childhood asthma. N Engl J Med 2011;364:701-9.

30. Strina A, Barreto ML, Cunha S, de Fátima SP, de Oliveira M, Moreira SC, et al. Validation of epidemiological tools for eczema diagnosis in Brazilian children: the ISAAC’s and UK Working Party’s criteria. BMC Dermatol 2010;10:11.

31. Amberbir A, Medhin G, Alem A, Britton J, Davey G, Venn A. The role of acetaminophen and geohelminth infection on the incidence of wheeze and eczema: a longitudinal birth-cohort study. Am J Respir Crit Care Med 2011;183:165-70.
| Variable | Overall | SPT reactivity to HDM | SPT reactivity to perennial allergens |
|----------|---------|-----------------------|-------------------------------------|
|          | No. (%) | Prevalence (%)        | Crude OR (95% CI) P value | Adjusted OR (95% CI) P value | Prevalence (%) | Crude OR (95% CI) P value | Adjusted OR (95% CI) P value |
| Any maternal geohelminth | | | | | | | |
| No | 1119 (54.1) | 10.2 | 1 | 1 | 13.9 | 1 | 1 |
| Yes | 950 (45.9) | 6.9 | 0.66 (0.48-0.91) .010 | 0.61 (0.43-0.86) .005 | 0.72 (0.55-0.95) .019 | 0.69 (0.51-0.92) .012 |
| Maternal age (y) | | | | | | | |
| ≤20 | 535 (25.9) | 8.0 | 1 | 1 | 12.1 | 1 | |
| 21-29 | 1002 (48.4) | 8.9 | 1.13 (0.77-1.67) .517 | | 12.5 | 1.03 (0.75-1.43) .837 |
| ≥30 | 532 (25.7) | 8.9 | 1.14 (0.73-1.75) .567 | | 12.2 | 1.00 (0.69-1.45) .982 |
| Maternal ethnicity | | | | | | | |
| Non–Afro-Ecuadorian | 1524 (73.7) | 8.5 | 1 | 1 | 10.5 | 1 | 1 |
| Afro-Ecuadorian | 545 (26.3) | 9.3 | 1.10 (0.78-1.55) .578 | 1.11 (0.77-1.59) .586 | 12.8 | 1.07 (0.79-1.43) .700 | 1.07 (0.79-1.46) .659 |
| Maternal educational level | | | | | | | |
| Illiterate | 317 (15.3) | 8.4 | 1 | 1 | 11.7 | 1 | 1 |
| Complete primary | 1203 (58.1) | 8.7 | 1.04 (0.66-1.63) .860 | 0.82 (0.50-1.34) .419 | 12.5 | 1.08 (0.74-1.60) .682 | 0.94 (0.56-1.57) .768 |
| Complete Secondary | 549 (26.6) | 8.7 | 1.04 (0.63-1.72) .879 | 0.58 (0.32-1.05) .070 | 12.2 | 1.06 (0.69-1.63) .798 | 0.73 (0.43-1.21) .221 |
| Area of residence | | | | | | | |
| Urban | 1458 (70.5) | 9.5 | 1 | 1 | 13.1 | 1 | 1 |
| Rural | 611 (29.5) | 6.8 | 0.70 (0.49-1.01) .055 | 0.83 (0.55-1.25) .363 | 10.5 | 0.78 (0.58-1.05) .105 | 0.85 (0.60-1.20) .347 |
| Sex | | | | | | | |
| Male | 1052 (50.9) | 9.0 | 0.93 (0.68-1.26) .629 | 0.92 (0.67-1.26) .618 | 11.7 | 0.89 (0.68-1.16) .383 | 0.88 (0.67-1.15) .346 |
| Female | 1017 (49.1) | 8.4 | 1 | 1 | 12.9 | 1 | 1 |
| Socioeconomic status* | | | | | | | |
| 1 | 678 (32.7) | 8.4 | 1 | 1 | 11.1 | 1 | 1 |
| 2 | 679 (32.8) | 6.7 | 1.36 (0.91-2.04) .133 | 1.32 (0.88-2.02) .181 | 12.2 | 1.12 (0.80-1.56) .508 | 1.11 (0.78-1.57) .653 |
| 3 | 712 (34.4) | 9.0 | 1.58 (1.07-2.33) .020 | 1.45 (0.93-2.26) .097 | 13.6 | 1.26 (0.91-1.74) .165 | 1.18 (0.82-1.71) .376 |
| Birth order | | | | | | | |
| First | 517 (25.0) | 10.4 | 1 | 1 | 13.6 | 1 | 1 |
| Second-fourth | 1148 (55.5) | 8.6 | 0.80 (0.57-1.15) .231 | 0.77 (0.54-1.12) .174 | 12.4 | 0.90 (0.66-1.23) .504 | 0.85 (0.62-1.17) .321 |
| ≥Fifth | 404 (19.5) | 6.8 | 0.63 (0.39-1.02) .058 | 0.61 (0.35-1.04) .070 | 10.6 | 0.75 (0.50-1.13) .172 | 0.70 (0.44-1.10) .125 |
| Maternal allergy | | | | | | | |
| No | 1959 (95.3) | 8.6 | 1 | 1 | 12.2 | 1 | 1 |
| Yes | 97 (4.7) | 10.4 | 1.24 (0.63-2.42) .538 | 1.33 (0.67-2.64) .422 | 15.6 | 1.34 (0.76-2.36) .318 | 1.40 (0.79-2.50) .250 |
| Household overcrowding† | | | | | | | |
| ≤3 | 1174 (56.7) | 8.9 | 1 | 1 | 12.1 | 1 | 1 |
| >3 | 895 (43.3) | 8.4 | 0.93 (0.68-1.27) .644 | 1.06 (0.81-1.38) .690 | 12.6 | 1.06 (0.81-1.38) .690 | 1.21 (0.90-1.63) .208 |
| Pets inside house | | | | | | | |
| No | 1544 (74.6) | 8.2 | 1 | 1 | 11.9 | 1 | 1 |
| Yes | 525 (25.4) | 10.0 | 1.24 (0.88-1.74) .882 | 1.15 (0.86-1.55) .355 | 13.5 | 1.15 (0.86-1.55) .355 | 1.19 (0.88-1.61) .260 |
| Large farm animals‡ | | | | | | | |
| No | 1405 (67.9) | 9.9 | 1 | 1 | 13.5 | 1 | 1 |
| Yes | 664 (32.1) | 6.5 | 0.63 (0.45-0.89) .009 | 0.72 (0.54-0.96) .025 | 10.1 | 0.72 (0.54-0.96) .025 | 0.76 (0.56-1.05) .095 |
| Pneumonia to 13 mo | | | | | | | |
| No | 1971 (95.5) | 8.9 | 1 | 1 | 12.5 | 1 | 1 |
| Yes | 94 (4.4) | 4.3 | 0.45 (0.16-1.25) .127 | 0.65 (0.31-1.36) .252 | 8.5 | 0.65 (0.31-1.36) .252 | 0.57 (0.28-1.26) .166 |
| Child geohelminths§ | | | | | | | |
| No | 1452 (71.1) | 9.1 | 1 | 1 | 12.9 | 1 | 1 |
| Yes | 590 (28.9) | 7.9 | 0.85 (0.60-1.21) .369 | 0.86 (0.64-1.59) .320 | 11.3 | 0.86 (0.64-1.59) .320 | 0.92 (0.66-1.26) .587 |

ORs and 95% CIs were estimated using logistic regression. P values of less than .05 are shown in boldface. Adjusted ORs are controlled for maternal ethnicity and educational level, area of residence, sex, socioeconomic status, birth order, maternal allergy, household overcrowding, pets inside the house, contact with large farm animals, pneumonia to 13 months, and geohelminths in the child.

* Socioeconomic status (SES) represents tertiles of z scores obtained by using a factor analysis, with 1 representing the lowest and 3 representing the highest SES.
† Household overcrowding is defined as the number of persons living in the household per sleeping room.
‡ Any of cows, pigs, mules, donkeys, and horses.
§ Geohelminth infections in the child: presence of any geohelminth infection detected during the first 2 years of life.
### TABLE E2. Crude associations between maternal geohelminth infections and study outcomes

| Geohelminth variable | Eczema to 3 y OR (95% CI) | P value | Wheeze to 3 y OR (95% CI) | P value | Allergen SPT reactivity at 3 y OR (95% CI) | P value | SPT reactivity to HDM at 3 y OR (95% CI) | P value |
|----------------------|---------------------------|---------|---------------------------|---------|------------------------------------------|---------|------------------------------------------|---------|
| Any maternal geohelminth presence |                          |         |                          |         |                                         |         |                                         |         |
| No (54.1%)           | 1                         | 1       |                          | 1       |                                         | 1       |                                         | 1       |
| Yes (45.9%)          | **1.31 (1.05-1.65)**      | **.018** | **1.17 (0.96-1.43)**     | **.110** | **0.84 (0.66-1.05)**                   | **.131** | **0.66 (0.48-0.91)**                   | **.010** |
| Maternal *A. lumbricoides* presence |                          |         |                          |         |                                         |         |                                         |         |
| No (72.5%)           | 1                         | 1       |                          | 1       |                                         | 1       |                                         | 1       |
| Yes (27.5%)          | **1.44 (1.13-1.84)**      | **.003** | **1.38 (1.11-1.72)**     | **.003** | **0.74 (0.57-0.97)**                   | **.011** | **0.51 (0.34-0.77)**                   | **.001** |
| Intensity            |                          |         |                          |         |                                         |         |                                         |         |
| Uninfected (77.8%)   | 1                         | 1       |                          | 1       |                                         | 1       |                                         | 1       |
| Light (17.1%)        | 1.28 (0.92-1.78)          | .150    | **1.33 (1.03-1.72)**     | **.027** | **0.68 (0.49-0.96)**                   | **.030** | **0.51 (0.31-0.84)**                   | **.008** |
| Moderate/heavy (5.1%)| 1.45 (0.85-2.50)          | .176    | **1.63 (1.07-2.48)**     | **.022** | **0.88 (0.52-1.51)**                   | .647    | **0.46 (0.18-1.15)**                   | .095    |
| Maternal *T. trichiura* presence |                          |         |                          |         |                                         |         |                                         |         |
| No (71.1%)           | 1                         | 1       |                          | 1       |                                         | 1       |                                         | 1       |
| Yes (28.9%)          | 1.21 (0.92-1.60)          | .181    | 1.16 (0.93-1.43)         | .181    | 0.85 (0.66-1.10)                       | .222    | **0.65 (0.45-0.94)**                   | **.022** |
| Intensity            |                          |         |                          |         |                                         |         |                                         |         |
| Uninfected (73.8%)   | 1                         | 1       |                          | 1       |                                         | 1       |                                         | 1       |
| Light (22.0%)        | 1.20 (0.88-1.64)          | .243    | 1.20 (0.88-1.64)         | .854    | 0.83 (0.62-1.11)                       | .209    | 0.66 (0.44-1.00)                       | .053    |
| Moderate/heavy (4.2%)| 1.75 (0.99-3.58)          | .053    | 1.75 (0.99-3.09)         | .576    | 0.67 (0.36-1.31)                       | .254    | **0.22 (0.05-0.92)**                   | **.037** |
| Maternal hookworm presence |                          |         |                          |         |                                         |         |                                         |         |
| No (94.4%)           | 1                         | 1       |                          | 1       |                                         | 1       |                                         | 1       |
| Yes (5.6%)           | 1.51 (0.92-2.49)          | .106    | 0.87 (0.56-1.36)         | .541    | 0.79 (0.46-1.35)                       | .384    | 0.79 (0.38-1.65)                       | .535    |

ORs and 95% CIs were estimated using logistic regression. P values of less than .05 are shown in boldface. Uninfected data for prevalence are derived from the results of all parasitologic methods, whereas those for infection intensity form Kato-Katz results.