

Ascariasis as a model to study the helminth/allergy relationships

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Summary

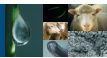
Ascariasis is the most frequent soil transmitted helminthiasis and, as well as other helminth infections, is expected to influence the clinical presentation of allergic diseases such as asthma. Indeed, several clinical and experimental works have shown an important impact either increasing or suppressing symptoms, and the same effects have been detected on the underlying immune responses. In this review we analyze the work on this field performed in Colombia, a Latin American tropical country, including aspects such as the molecular genetics of the IgE response to *Ascaris*; the allergenic activity of *Ascaris* IgE-binding molecular components and the immunological and clinical influences of ascariasis on asthma. The analysis allows us to conclude that the impact of ascariasis on the inception and evolution of allergic diseases such as asthma deserves more investigation, but advances have been made during the last years. The concurrent parasite-induced immunostimulatory and immunosuppressive effects during this helminthiasis do modify the natural history of asthma and some aspects of the practice of allergology in the tropics. Theoretically it can also influence the epidemiological trends of allergic diseases either by its absence or presence in different regions and countries.

1 | INTRODUCTION

For justified reasons, helminth parasites are considered detrimental organisms that, in addition to their well-known deleterious effects on millions of children, are able to manipulate the host immune system, stimulating or suppressing its response. The increasing knowledge of this dual effect has explained some (and raised many more) fundamental questions about the parasite/allergy relationships. In addition, there is a growing discovery of potential anti-inflammatory helminth products, positioning these parasites as protagonists in the current debate about the high prevalence and possible therapies of chronic non-communicable inflammatory diseases. With regard to allergy, there are scientific questions that could be explored through these relationships; for example, the origins of allergenicity, the mechanisms of allergenic activity and the evolutionary relationship between parasite immunity and allergy; and also practical questions such as the impact of cross-reactivity on the diagnosis of allergy.

Among the environmental factors influencing the pathogenesis, clinical manifestations and epidemiologic trends of allergic diseases in the tropics, there are two that seem to be the most important: permanent exposure to house dust mite (HDM) allergens (mainly *Blomia tropicalis* and *Dermatophagoides pteronyssinus*) and helminth infections.¹ At the Institute for Immunological Research (<http://www.worldallergy.org/wao-centers-of-excellence>) in Cartagena, a tropical Caribbean city of Colombia, we have analysed the relationships between ascariasis and asthma, as a model to understand several aspects of the helminth/allergy relationships.

Colombia is a tropical country with almost 50 million inhabitants, 79% of them living in urban areas, where the basic health indicators of the last two decades reflected a global and health improvement.² The country has a distinct regional climate; in the Caribbean Coast, the mean annual temperature is 28°C and the humidity 85%, whilst in the Andeans, they are, respectively, 12°C and 65% humidity. The prevalence of parasite infections (among them ascariasis, caused by the helminth *Ascaris lumbricoides*) ranges from 5% in urban centres³



to 50% in some rural communities,⁴ although the actual frequency could be higher as detected by serology.⁵ The national prevalence of allergic diseases has changed in 10 years from 12% to 12.5%,^{6,7} the increase being driven by the rates in children.

Our work has followed three major lines of research: the evolutionary links between the IgE responses to *Ascaris* and HDM allergens; the allergenic activity and cross-reactivity of *Ascaris* molecular components and the immunological and clinical influences of ascariasis on asthma. Here, we analyse data obtained from them and present some of the guiding hypotheses. Since ascariasis is the most frequent soil-transmitted helminthiasis, research about its impact on allergy is expected to be very instructive, but the role of other parasites should be kept in mind.

2 | GENETICS OF THE IGE RESPONSES TO ASCARIS AND HOUSE DUST MITE ALLERGENS

The polygenic character of immune responses to infective organisms has been supported in the last decades by a number of improvements on sequencing-based genotyping and genomewide transcriptional profiling. There is no need to explain the importance of finding genetic markers of resistance to helminthiasis or those that influence the specific IgE hyper-responsiveness to allergens. However, there are other aspects less evident such as the origin of allergenicity, which is the ability of a molecule to induce the production of specific IgE. Studying this in helminth models, especially in the tropics where high and low responders can be easily detected, would help to understand basic mechanisms of allergenicity and allergenic activity, the latter being the ability of specific IgE to induce an inflammatory cascade and clinical symptoms.

Besides, there is a more general topic: the evolutionary relationship between parasite immune responses and immune mechanisms of allergy, a field with several hypotheses that can be advantageously tested in the tropics. In this regard, we have remarked that a fundamental unresolved question is whether current genotypes predisposing to allergic response were selected by helminth infections or, to the contrary, these genotypes (shaped by factors different to parasites) generated resistance and exerted a selective pressure on them, in such a way that those with better immunomodulatory capacity survived as parasites.^{8,9} This possibility seems to be more in agreement with a general basis of parasitism, which considers that the success of parasitic relationships depends on both the parasite virulence and the host resistance (Figure 1). Resistance to helminths involves several innate and adaptive mechanisms, including antibody responses, whose protective role varies among parasite species, but independently of this effect, specific IgE hyper-responsiveness is a complex trait whose relationship with allergy to common allergens could be better analysed in places co-exposed to both sources; therefore, our work has been focused on these aspects.

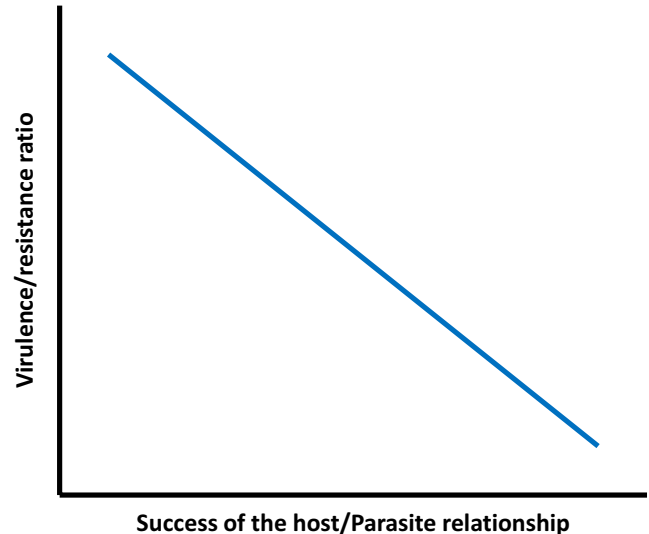


FIGURE 1 Factors determining the success of the parasitism relationships. The survival of both host and parasite increase if parasite virulence or host resistance decreases as shown in the crossline. One of the reasons the parasite virulence can decrease is the natural selection of those with greater immunomodulatory properties; which in turn modify the immune responses, including allergy inflammation. There is also evidence that humans with strong type-2 immunity to helminths limit the parasitism relationship and promote allergic responses

The genetic variants influencing antibody responses to *A. lumbricoides* are still unclear. After the discovery of linkage of egg counts and total IgE with two loci in human chromosomes 1 and 13 by Williams-Blangero et al,¹⁰ our group detected a polymorphism in the gene encoding the B-cell activating factor (Chr. 13q33) associated with levels of specific IgG to *Ascaris*.¹¹ The potential functional effect of this locus was further suggested by the inverse relationship between BAFF levels (protein and mRNA) and antibody levels to *Ascaris* in humans.¹² In addition, by targeted resequencing of 14 immune gene regions, we recently detected 70 genetic variants distributed in eight genes that were enriched in high IgE responders to the whole *Ascaris* extract and to the *Ascaris* resistance marker ABA-1. We found associations between the IgE response to *Ascaris* and the chitinase-related genes acidic chitinase (CHIA) and chitinase 3-like 1 (CHI3L1) in Chr. 1, as well as Chr. 13q33-34 ligase IV (LIG4) and abhydrolase domain containing 13 (ABHD13).¹³

Chitinase-related genes acidic chitinase encodes acidic mammalian chitinase (AMCase), an enzyme with functional catalytic and chitin-binding domains in humans; it stimulates IL-13 production¹⁴ and some variants have been previously associated with asthma.¹⁵ We also found that polymorphism rs4950928 CHI3L1 was associated with high IgE levels to ABA-1 in the Colombian population and IgE levels to the birch pollen allergen Bet v 1 in a Swedish cohort unexposed to *Ascaris*.¹³ Together, these findings suggest that chitinase-related genes might be involved in immune responses to both *Ascaris* and non-parasitic allergens and act as a marker of pro-Th2 responses.

In regard to total IgE, our results after fine mapping the 13q33 region showed significant associations between two polymorphisms in TNFSF13B encoding the cytokine B-cell activating factor (BAFF) with increased total IgE levels in the Colombian and Swedish population; in addition, a significant association between a variant of insulin receptor substrate 2 (IRS2) and increased IgE levels was found. IRS-2 is an intracellular adaptor that is phosphorylated after interleukin 4-receptor engagement and participates in receptor tyrosine kinase signalling leading to IgE production.¹⁶ Then, after its discovery,¹⁰ we have added data supporting the idea that the 13q33 locus influence IgE levels in humans. Indeed, in ongoing analyses, we have found associations between TNFSF13B variants, the expression of its mRNA transcripts and levels of specific IgE to *Ascaris* extract (manuscript in preparation).

Our studies include the simultaneous evaluation of the IgE antibody response to *Ascaris* and HDM allergens among asthmatics and healthy controls from the same population, which has been extensively characterized as an admixture of Spaniards, African and Native Americans.¹⁷ Most of the associations with IgE to *Ascaris* components were not found with HDM allergens, and there was no association of these variants with asthma in our population. These findings do not support the hypothesis that IgE specific to HDM allergens is a consequence of protective immunity to helminths. However, the detection of a CHI3L1 variant associated with the IgE response to both ABA-1 and Bet v 1 does support it. Thus, more studies involving large populations and the whole genome are needed to get a better picture of the evolutionary relationships between helminth immunity and the allergic responses, which is largely dependent on the general parasitism relationships (Figure 1).

3 | THE ALLERGENIC ACTIVITY OF ASCARIS IGE-BINDING MOLECULAR COMPONENTS

In general, helminths have IgE-binding molecules that in some cases behave as truly allergens.¹⁸ In the particular case of *Ascaris*, its allergenic activity is evident during the natural infection in humans^{19–22} and also in experimental models in primates^{23,24} and mice.²⁵ In addition, pig models of sensitization and challenge with *Ascaris* extracts have shown asthma symptoms.^{26,27} Furthermore, epidemiologic surveys have reported positive association between active infection and asthma symptoms.²⁸ These pulmonary manifestations are caused by the larvae migration through the lungs, which induces an IgE-mediated inflammatory response, suggesting that *Ascaris* has great allergenic properties. However, although several IgE-binding molecules are present in the whole-body extract, few of them have been characterized as real allergens. Therefore, in addition to their capacity to induce and bind human IgE, we are currently evaluating the allergenic activity of some *Ascaris* molecular components. Three *A. lumbricoides* allergens are listed in the official WHO/IUIS site, one species-specific (Asc s 1, also known as ABA-1) and two cross-reacting, Asc I 3 (tropomyosin) and Asc I 13

(glutathione-S-transferase), although at least nine additional IgE-binding components have been reported.²⁹

3.1 | Asc s 1

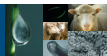
This molecule was first identified in *A. suum* but is also present in *A. lumbricoides*. It is officially accepted by the IUIS as an allergen because it binds human IgE, but its allergenic activity remains to be demonstrated. In humans, the IgE and IgG responses to ABA-1 have been associated with protection from ascariasis³⁰ rather than allergy symptoms. Indeed, ABA-1 is a species-specific antigen and is considered a nematode-specific infection-marker; it does not cross-react with any of the *B. tropicalis* or *D. pteronyssinus* allergens.²⁹ Studies in different populations show frequencies of sensitization from 25% to 70%,^{31,32} reaching 75% in *Ascaris*-sensitized children,⁵ which is expected because the pseudocelomic fluid of *Ascaris* contains abundant amounts of this polyprotein.³³ However, the relationship of this response with asthma is not clear; for example, in one study, we found no association between asthma and sensitization to Asc s 1,³⁴ but another report showed a strong relationship with the severity of asthma symptoms.³¹ ABA-1 belongs to a family of fatty acid binding proteins only found in nematodes; it binds different lipids and retinoids, but the influence of these biological properties on its allergenicity is unknown.^{35,36}

3.2 | Asc I 3

Ascaris tropomyosin has strong cross-reactivity with those present in HDM and cockroaches, which is explained by the high structural homology between them.^{29,37,38} Tropomyosin is the most abundant protein of the worm's body; thus, specific IgE highly contributes to the IgE levels detected with the somatic extract. It is one of the best-characterized allergens from this helminth; there is evidence that it may induce mast cell degranulation and basophil histamine release.⁸ Sensitization to this allergen is associated with asthma presentation and severity in the Colombian population^{31,34}; in addition, because of its high cross-reactivity, an influence on allergy diagnosis, including that of food allergy, is expected.³⁹

3.3 | Asc I 13

The glutathione-S-transferase (GST) family from *Ascaris* has different isoforms with IgE-binding capacity; however, a member of the sigma class (Asc I 13) has been studied as a purified molecule. The frequency and strength of sensitization in the general population is low, and natural GST induces weak positive skin test responses in asthmatic patients.⁴⁰ Antibody responses to *Ascaris* GST could be particularly important in the first years of life when *Ascaris* immunity starts. In early childhood, it is the most frequent *Ascaris* IgE-binding component, inducing also the greatest specific titres⁴¹; however, this response decreases with age⁴⁰ and there is no association with asthma.³⁴ The cross-reactivity of Asc I 13 with its orthologue in HDM is still a matter of controversy. Based on a proteomic



approach, cross-reactivity with the GST from *B. tropicalis* was found; however, there was no confirmation when testing the purified recombinants Asc I 13 and Blo t 8 (the *B. tropicalis* GST) at least in the North American population.⁴² Homology of amino acid sequence between Asc I 13 and Blo t 8 is moderate, but IgE cross-reactivity has been found between the phylogenetically distant filarial and cockroach GSTs⁴³ that share intermediate similarity. It is possible that other isoforms of GST mediate cross-reactivity between these species because at least six natural isoforms are expected in each organism.⁴⁰ Public databases of *Ascaris* genome report five GST sequences with different predicted isoelectric points and belonging to several classes. We have confirmed the IgE-binding properties of an Omega GST isoform from *Ascaris* with an enzymatic activity different from that of the sigma GST. Polyclonal antibodies from Asc I 13 immunized animals do not recognize this new isoform, suggesting limited cross-reactivity.⁴⁴ An extensive characterization of the allergenic properties of members of *Ascaris* GST family is an unmet need.

3.4 | AI16

We isolated a clone coding for AI16 by IgE immunoscreening of an *A. lumbricoides* cDNA library using sera from parasitized children. The protein is almost identical to As16, an antigen of *A. suum* that has been characterized in the race of developing vaccines against ascariasis. The recombinant version of AI16 has strong and frequent IgE-binding activity: 53.3% in normal children and 65.5% in asthmatic patients sensitized to *Ascaris*, both groups living in economically deprived communities in Cartagena. Also, it induces basophil activation and histamine release in cells from individuals living in endemic villages and also passive cutaneous anaphylaxis in BALB/mice (manuscript in preparation). Another *A. lumbricoides* IgE-binding molecule, this with low allergenic activity, is cystatin, which has been studied as an immunomodulator (see below). The reasons why some *Ascaris* components that induce and bind IgE exhibit different degrees of allergenic activity are a matter of active research in our laboratory.

4 | IMMUNOLOGICAL AND CLINICAL INFLUENCES OF ASCARIASIS ON ASTHMA

A major challenge for Latin American groups working on allergic diseases was to scientifically document the high prevalence of allergic respiratory symptoms in the region and the tropics.¹ The obvious, still open question is why in developing countries, with limited hygiene conditions, the prevalence of wheezing and asthma is as high as that observed in wealthy societies. As possible explanations, we have proposed the permanent exposure to mite allergens⁴⁵⁻⁴⁷ and helminth infections,^{5,8,34,48} although genetic factors may be involved.⁴⁹⁻⁵¹ However, the question does not necessarily have to be raised within the framework of hygiene hypothesis because other, more general factors, such as biodiversity and climate change could be involved with different relative role in geographic zones.^{52,53}

Besides, there are diagnostics problems that could limit the accuracy of some surveys, especially in children where viral respiratory infections are a frequent cause of wheezing.^{1,54,55}

4.1 | The role of HDM permanent exposure

The proposed role of HDM in the increasing prevalence of asthma in industrialized countries^{56,57} is also valid, with even more reasons, in tropical regions where perennial exposure induces high frequency of sensitization with more clinical impact than other allergens.^{58,59} It has been reported that maternal HDM exposure during pregnancy enhances severity of HDM-induced asthma in murine offspring in an antigen-specific way, which was associated with greater Th2 cytokine production, frequency of IL-13-producing T cells in the lung and immunoglobulin production.⁶⁰ Theoretically, these findings can occur in humans, supporting a role of permanent HDM exposure on the inception of asthma; therefore, this should be further investigated, especially because negative results have been obtained when exploring human cord blood cell proliferation in similar conditions.⁶¹

Several studies have shown that HDM sensitization is the main risk factor for asthma in the tropics (reviewed in Refs [1,62]); but its historic epidemiological impact is difficult to evaluate because scientific reports on asthma prevalence in tropical countries are relatively new, which makes it impossible to determine whether the current rates reflect a recent increase or a more permanent phenomenon; although mite levels of exposure might be modified by urbanization, climate change and Westernized lifestyles.

4.2 | The role of helminthiasis

It has become progressively clear that helminthiasis may influence several aspects of allergy, such as prevalence, diagnosis and severity ([63-65] reviewed in Ref. [1]) Ascariasis may increase allergic sensitization⁶⁶ and asthma symptoms in different ways, among them allergen cross-reactivity and nonspecific boosting of mite-specific IgE responsiveness ([67]; experimental work also reviewed in Ref. [68,69]). Under appropriate conditions (eg, low-intensity infections as occurs in urban settings), these factors overshadow the immune suppressor effects, probably leading to the positive associations between asthma and helminthiasis reported by several groups⁷⁰⁻⁷² (reviewed in Ref. [69]).

4.3 | Cross-reactivity between HDM and *Ascaris* components

Due to the permanent co-exposure to HDM and *Ascaris*, it is expected that the IgE responses to common epitopes from both sources could be increased, which has been supported by some experimental works.^{73,74} There is high cross-reactivity between HDM and *Ascaris*, in part determined by tropomyosins and GSTs, but other non-characterized components are also involved. We evaluated the role of these and other allergens as risk factors for asthma presentation and severity. In a nationwide study including 356 asthmatics

and 435 controls, it was confirmed that sensitization to both *Ascaris* and *D. pteronyssinus*, as well as their tropomyosins, was risk factors for asthma,³⁴ showing for the first time a molecular component of *Ascaris* strongly associated with this disease. In another asthma cohort, it was also found that not only sensitization to *Ascaris* extract but also to HDM and *Ascaris* components were associated with symptoms of asthma severity.³¹ It has been proposed that patients genetically predisposed to asthma, early age parasitized, under deworming programs, suffering several re-infections and permanently exposed to HDM probably have a stronger IgE response to allergens and more severe clinical symptoms. Indeed, several works show a role of *Ascaris* sensitization to species-specific and cross-reactive allergens in asthma pathogenesis^{34,75,76} and together with other studies⁷⁷⁻⁷⁹ support that ascariasis is an important factor modifying the IgE responses and the natural history of asthma in the tropics.

It may be worth noting that IgE-binding carbohydrate determinants could be implicated in cross-reactivity between helminth and HDM; in fact, a galactose β 1-4fucose epitope has been detected in N-glycans shared by *A. suum* and *D. pteronyssinus*.⁸⁰ We have found several glycosylated components of high molecular weight (>100 kDa) in *A. lumbricoides* extract that were inhibited by pre-absorption of sera with HDM extracts; however, deglycosilation does not significantly affect the specific IgE antibody binding to the extract.²⁹ Cross-reactivity due to carbohydrate determinants may affect specific IgE measurements in vitro.⁸¹ For example, a limitation of serologic allergy diagnosis in patients with helminthiasis could be the presence of the carbohydrate epitope galactose- α 1,3-galactose (α -Gal), expressed in non-primate mammalian proteins.⁸² However, the presence of this epitope in *Ascaris* spp. has not been demonstrated.

4.4 | Nonspecific boosting of the immune responses

Theoretically, the strong Th2 responses during ascariasis could nonspecifically increase the memory responses to non-related allergens. However, it is still a mystery what components of *Ascaris* are the inducers of this type of response that, at the end, might favour parasitism⁸³ and no "sterile immunity." There are few reports showing experimentally that specific *Ascaris* molecules induce a Th2 response,^{84,85} although indirect evidence shows that it has several IgE inducers, such as carbohydrates,⁸⁶ glycolipids⁸⁷ and the proteins ABA-1, tropomyosin and GST.^{29,30,38,40} Whether these components are genuinely responsible for the Th2 responses or there are non-specific Th2 inducers that create an appropriate environment for specific IgE responses⁸⁸ remains to be defined, but there is evidence that ascariasis can boost the IgE/Th2 responses to bystander antigens.⁸⁹⁻⁹¹ Since it might help to explain the allergenic effects of *Ascaris*, we have explored this aspect in human ascariasis.

Evaluating the evolution of the immune responses in children of the FRAAT birth cohort,³ we observed that *Ascaris*-sensitized children had stronger IgE to *B. tropicalis*. To rule out the effect of cross-reactivity, species-specific components were used, finding that those reacting with IgE to ABA-1 had increased IgE responses to Blo

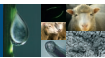
t 5 and Blo t 12, two species-specific allergens of *B. tropicalis*.⁹² This *Ascaris*-induced nonspecific boosting was also detected in a large nationwide population co-exposed to *A. lumbricoides* and HDM, where IgE levels and frequencies of sensitization to mite allergen extracts were greater in subjects sensitized to ABA-1. In fact, these individuals had at least twice the odds of being sensitized to HDM.³⁴ Furthermore, in a cohort of asthmatic patients living in the tropics, *Ascaris*-sensitized patients had significantly higher IgE levels to the HDM allergens Der p 2 and Blo t 5.³¹ All these findings support the idea that the Th2/IgE hyper-responsiveness induced by *Ascaris* infection includes the responses not only to *Ascaris* antigens but also to HDM allergens. It is well known that ascariasis induces a polyclonal nonspecific stimulation of B cells⁶⁷; therefore, it can be hypothesized that the involved components also stimulate mite allergen-specific memory B cells that in the tropics are theoretically in permanent allergen-specific stimulation. Since specific IgE to HDM is the most important risk factor for asthma in the tropics, any condition that increases the allergic response against them could also increase symptoms and severity of this disease.^{31,65} *Ascaris* components and host genetic background influencing this boosting effect are only beginning to be unravelled, deserving further investigations.

4.5 | Cross-reactivity and allergy diagnosis

Helminthiasis jeopardize the diagnosis of allergy not only because sometimes they induce allergy symptoms but also because the similar immune responses can confound laboratory parameters such as total IgE and blood/sputum eosinophilia (reviewed in Ref. [9]) and (theoretically) other eosinophil-related biomarkers such as exhaled nitric oxide and eosinophil cationic protein. Under these circumstances, specific IgE remains the most appropriate biomarker for allergy diagnosis in the tropics, although in some cases the cross-reactivity between HDM and *Ascaris* components can interfere the serological diagnosis.

In the majority of cases, skin testing with mite extracts can detect sensitization in patients with asthma, being component-resolved diagnosis very useful when genuine sensitization is not clear. Component-resolved decision algorithms for both temperate and tropical countries have been proposed,⁹³ considering cross-reactivity between mite tropomyosins (Blo t 10 and Der p 10) and *Ascaris* tropomyosin (Asc I 3) as a diagnostic confounder. However, as already mentioned there is evidence that in the tropics IgE to *Ascaris* tropomyosin could influence asthma symptoms,³⁴ so more studies are needed to define its impact on respiratory allergy, especially because we have evidence that tropomyosins from other sources such as mosquitoes induce IgE sensitization.^{94,95}

Although cross-reactivity between HDM (Der p 8, Blo t 8), cockroach (Bla g 5) and *Ascaris* GST (Asc I 13) has been detected using sera from individuals living in the tropics,⁴⁰ it seems that that observed between cockroach and Asc I 13 is not a problem in the United States.⁴² Cross-reactivities of HDM (Blo t 10) and mosquito tropomyosin (Aed a 10) with shrimp tropomyosin Lit v 1 have been described.⁹⁴ Asc I 3 shares epitopes with shrimp tropomyosins;



therefore, cross-reactive IgE response is expected and could interfere with the serological diagnosis of allergy to shellfish tropomyosin. Then, in the tropics, there are four known sources of tropomyosins (helminths, HDM, shellfish and mosquitoes) that induce IgE sensitization, which precludes their use as specific markers for allergy diagnosis. This could be overcome if epitope specific diagnosis becomes available, but the effect of the allergenic burden and particularly that of helminth tropomyosins on the inception of food tropomyosin allergy remains to be studied.

4.6 | The patterns of IgE sensitization to HDM and *Ascaris* in humans

It is generally accepted that patterns of sensitization and risk for allergic diseases depend on the genetic background and early-life environmental exposure.^{3,92} The IgE responses to *Ascaris* and HDM grow in parallel since early life in co-exposed tropical inhabitants.⁵ Results obtained from longitudinal analyses of IgE to *Ascaris* and *B. tropicalis* in a birth cohort of children living in Cartagena, Colombia (FRAAT study) indicate a high correlation between them, and a half of *Ascaris*-sensitized children also respond to HDM. Remarkably, almost a third of the 3-year-old cohort showed positive *B. tropicalis* responses independently of presenting allergic symptoms.

Using specific markers for both sources *Asc* s 1, *Blo* t 5 and *Blo* t 12,^{96,97} primary sensitization to both *B. tropicalis* and *Ascaris* was found.⁵ Almost 25% of children as young as 6 months were sensitized to *Asc* s 1 and the cross-reactive *Asc* l 3 and *Asc* l 13. The IgE antibodies to *Asc* l 3 detected before the first year of life highly correlated with those against *Asc* s 1 and *Asc* l 13 but not with those to the *B. tropicalis* components *Blo* t 5 and *Blo* t 12, suggesting that the cross-reactive anti-tropomyosin IgE response is primarily induced by *Asc* l 3.⁴¹ Regarding the environmental factors influencing the IgE responses to *Ascaris* and HDM, poverty and unhygienic living conditions increase the odds of sensitization for both sources in children.⁵ The reason for this pattern seems to be beyond cross-reactivity because we have also observed that poverty is a risk factor for sensitization to the HDM specific markers *Blo* t 5 and *Blo* t 12 (JZ, NA, LC, unpublished data).

4.7 | *Ascaris*-induced immunoregulation

Ascaris spp. are also able to down-regulate the immune responses of hosts,⁹⁸ which explains why it is one of the most successful parasites (Figure 1). This has been shown in both, humans and experimental animals, using various strategies. For example, different patterns of cytokine production have been detected among young adults living in an *A. lumbricoides* endemic area. Chronically infected *Ascaris* and *Trichuris* patients with high parasite load presented reduced cellular reactivity and lower type 1 cytokines TNF- α , IFN- γ and IL-12 than those non-infected endemic controls.⁹⁹ This immune hypo-responsiveness has been associated with greater frequencies of the spontaneous production of IL-10¹⁰⁰ and modified Th2-like immune phenotype¹⁰¹ that is also able to impair the immune response

to co-infecting parasites¹⁰² and probably the normal response to vaccines.

Whether *Ascaris*/*Trichuris*-induced immune hypo-responsiveness has an impact on preventing asthma or other allergic diseases remains to be defined, but recent reports show that soil-transmitted helminthiasis during childhood protect from wheezing and asthma but not from allergen sensitization.^{86,103} Since *Ascaris* and *Trichuris* co-infection is very frequent, it is difficult to evaluate their individual down-regulatory properties in humans; however, progress on the molecular characterization of parasite immunomodulatory components is expected to facilitate this research.

The mechanisms of the immune down-regulatory effects of *A. suum* infection have been recently explored in a LPS-induced inflammatory response in mice,¹⁰⁴ finding that infection suppressed secretion of pro-inflammatory cytokines (IL-1 β , TNF- α and IL-6) and induces high levels of IL-10 and TGF- β , as well as CD4⁺CD25^{high}, Foxp3⁺ T cells in the mesenteric lymph nodes. This has also investigated in pigs using transcriptomic analysis, showing that chronic *A. suum* infection induces suppression of inflammatory pathways in the intestinal mucosa, down-regulating genes encoding cytokines, as well as antigen-processing and co-stimulatory molecules. The effect was reproduced by *A. suum* body fluid in human dendritic cells (DCs) in vitro.¹⁰⁵

The immunomodulatory capacity of *A. suum* and *A. lumbricoides* components has also been studied in animal models,¹⁰⁶⁻¹⁰⁸ and some immunomodulatory components found in excretory/secretory products of *Ascaris* have been identified. Among them, phosphorylcholine-containing glycosphingolipids^{109,110} and PAS-1^{84,111} have been more analysed; however, according to the *A. suum* genome,¹¹² this genus might have more than 15 potential immunomodulators. In this regard, the recent analysis of the anti-inflammatory properties of *A. lumbricoides* cystatin (rAI-CPI)¹¹³ started a new stage of research on the immunosuppressive face of this nematode. Cystatins, a family of cysteine protease inhibitors, are well-known immunosuppressors and have been identified in several helminths such as *O. volvulus*, *B. malayi*, *N. brasiliensis* and *A. viteae*; that from the last species (AvCystatin) being the most studied.¹¹⁴

The rAI-CPI was able to ameliorate the inflammation in a mouse model of DSS-induced colitis. Treatment with this molecule significantly reduced disease activity index, myeloperoxidase activity and inflammation score without toxic effects. Also, IL-10 and TGFB gene overexpression was observed together with a reduction in IL-6 and TNFA expression. rAI-CPI also induced the production of IL-10 and TGFB by macrophages,¹¹³ and further work in an animal model of airway inflammation has confirmed its IL-10-associated anti-inflammatory activity showing the induction of regulatory T cells as well as the production of IL-10 by human monocyte-derived dendritic cells.¹¹⁵ In addition, since some helminth products could be potential inducers of adverse allergic reactions, several tests for allergenic activity have been done on rAI-CPI, finding that it binds IgE but does not have allergenic activity¹¹⁶; therefore, this *A. lumbricoides*-derived immunomodulator seems promising for treating immune-mediated inflammation of intestine and lung.

Since *Ascaris* have both stimulating and suppressing effects on the Th2 responses, an important unsolved question is why one of them predominates during infections. As mentioned before, several studies suggest that light infections promote the Th2 responses and chronic severe infections suppress it. In the tropics, the later are present in rural areas, whilst intermittent and low-intensity infections occur mainly in urbanized areas. Therefore, a gradient of *A. lumbricoides* effects on the allergic response can be observed in this zone.⁵³ Due to the increasing trends of urbanization, the stimulating effects on the Th2 responses are currently more frequent, which is just a historical circumstance and does not mean that *Ascaris* is unable to induce immunosuppression, a property that seems to be important for surviving as parasite (Figure 1).

5 | CONCLUSIONS

The impact of ascariasis on the inception and evolution of allergic diseases such as asthma is part of the long co-evolutionary history of helminths and humans and deserves more investigation, but advances have been made during the recent years. The concurrent parasite-induced immunostimulatory and immunosuppressive effects during ascariasis can modify the natural history of asthma and some aspects of the practice of allergology in the tropics. Theoretically, it can also influence the epidemiological trends of allergic diseases either by its absence or presence in different regions and countries. Basic research in this field will help to understand fundamental questions about parasite immunity and allergy.

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REFERENCES

- Caraballo L, Zakzuk J, Lee BW, et al. Particularities of allergy in the Tropics. *World Allergy Organ J*. 2016;9:20.
- OPS (2017) Salud en las Américas, Edición del 2017. Resumen: panorama regional y perfiles de país. OPS Publicación Científico Técnica No 642: 111-114.
- Acevedo N, Sanchez J, Zakzuk J, et al. Particular characteristics of allergic symptoms in tropical environments: follow up to 24 months in the FRAAT birth cohort study. *BMC Pulm Med*. 2012;12:13.
- Agudelo-López S. Prevalencia de Parasitosis Intestinales y Factores Asociados en un Corregimiento de la Costa Atlántica Colombiana. *Rev Salud Publica*. 2008;10:633-642.
- Zakzuk J, Acevedo N, Cifuentes L, et al. Early life IgE responses in children living in the tropics: a prospective analysis. *Pediatr Allergy Immunol*. 2013;24:788-797.
- Dennis R, Caraballo L, Garcia E, et al. Asthma and other allergic conditions in Colombia: a study in 6 cities. *Ann Allergy Asthma Immunol*. 2004;93:568-574.
- Dennis RJ, Caraballo L, Garcia E, et al. Prevalence of asthma and other allergic conditions in Colombia 2009-2010: a cross-sectional study. *BMC Pulm Med*. 2012;12:17.
- Acevedo N, Caraballo L. IgE cross-reactivity between *Ascaris lumbricoides* and mite allergens: possible influences on allergic sensitization and asthma. *Parasite Immunol*. 2011;33:309-321.
- Caraballo L. *Ascaris* and Allergy. In: Holland CV, ed. *Ascaris: the neglected parasite*. London, UK: Elsevier; 2013:21-50.
- Williams-Blangero S, VandeBerg JL, Subedi J, et al. Genes on chromosomes 1 and 13 have significant effects on *Ascaris* infection. *Proc Natl Acad Sci USA*. 2002;99:5533-5538.
- Acevedo N, Mercado D, Vergara C, et al. Association between total immunoglobulin E and antibody responses to naturally acquired *Ascaris lumbricoides* infection and polymorphisms of immune system-related LIG4, TNFSF13B and IRS2 genes. *Clin Exp Immunol*. 2009;157:282-290.
- Bornacelly A, Mercado D, Acevedo N, Caraballo L. The strength of the antibody response to the nematode *Ascaris lumbricoides* inversely correlates with levels of B-Cell Activating Factor (BAFF). *BMC Immunol*. 2014;15:22.
- Acevedo N, Bornacelly A, Mercado D, et al. Genetic Variants in CHIA and CHI3L1 Are Associated with the IgE Response to the *Ascaris* Resistance Marker ABA-1 and the Birch Pollen Allergen Bet v 1. *PLoS ONE*. 2016;11:e0167453.
- Zhu Z, Zheng T, Homer RJ, et al. Acidic mammalian chitinase in asthmatic Th2 inflammation and IL-13 pathway activation. *Science*. 2004;304:1678-1682.
- Chatterjee R, Batra J, Das S, Sharma SK, Ghosh B. Genetic association of acidic mammalian chitinase with atopic asthma and serum total IgE levels. *J Allergy Clin Immunol*. 2008;122:202-208. 208 e201-207.
- Kelly-Welch AE, Wang HY, Wang LM, et al. Transgenic expression of insulin receptor substrate 2 in murine B cells alters the cell density-dependence of IgE production in vitro and enhances IgE production in vivo. *J Immunol*. 2004;172:2803-2810.
- Mathias RA, Taub MA, Gignoux CR, et al. A continuum of admixture in the Western Hemisphere revealed by the African Diaspora genome. *Nat Commun*. 2016;7:12522.
- Caraballo L, Coronado S. Parasite allergens. *Mol Immunol*. 2018;100:113-119.
- Löffler W. Transient lung infiltrations with blood eosinophilia. *Int Arch Allergy Appl Immunol*. 1956;8:54-59.
- Spillmann RK. Pulmonary ascariasis in tropical communities. *Am J Trop Med Hyg*. 1975;24:791-800.
- Gelpi AP, Mustafa A. Seasonal pneumonitis with eosinophilia. A study of larval ascariasis in Saudi Arabs. *Am J Trop Med Hyg*. 1967;16:646-657.
- Ribeiro JD, Fischer GB. Eosinophilic lung diseases. *Paediatr Respir Rev*. 2002;3:278-284.
- Patterson R, Harris KE, Pruzansky JJ. Induction of IgE-mediated cutaneous, cellular, and airway reactivity in rhesus monkeys by *Ascaris suum* infection. *J Lab Clin Med*. 1983;101:864-872.
- Patterson R, Harris KE. IgE-mediated rhesus monkey asthma: natural history and individual animal variation. *Int Arch Allergy Immunol*. 1992;97:154-159.
- Enobe CS, Araujo CA, Perini A, et al. Early stages of *Ascaris suum* induce airway inflammation and hyperreactivity in a mouse model. *Parasite Immunol*. 2006;28:453-461.
- Sylvain H, Dahlback M, Van Der Ploeg I, Alving K. The tryptase inhibitor APC-366 reduces the acute airway response to allergen in pigs sensitized to *Ascaris suum*. *Clin Exp Allergy*. 2002;32:967-971.

27. Thomas DJ, Husmann RJ, Villamar M, et al. *Lactobacillus rhamnosus* HN001 attenuates allergy development in a pig model. *PLoS ONE*. 2011;6:e16577.
28. da Silva ER, Sly PD, de Pereira MU, et al. Intestinal helminth infestation is associated with increased bronchial responsiveness in children. *Pediatr Pulmonol*. 2008;43:662-665.
29. Acevedo N, Sánchez J, Erler A, et al. IgE cross-reactivity between *Ascaris* and domestic mite allergens: the role of tropomyosin and the nematode polyprotein ABA-1. *Allergy*. 2009;64:1635-1643.
30. McSharry C, Xia Y, Holland CV, Kennedy MW. Natural immunity to *Ascaris lumbricoides* associated with immunoglobulin E antibody to ABA-1 allergen and inflammation indicators in children. *Infect Immun*. 1999;67:484-489.
31. Buendía E, Zakzuk J, Mercado D, Alvarez A, Caraballo L. The IgE response to *Ascaris* molecular components is associated with clinical indicators of asthma severity. *World Allergy Organ J*. 2015;8:8.
32. Turner JD, Faulkner H, Kamgno J, et al. Allergen-specific IgE and IgG4 are markers of resistance and susceptibility in a human intestinal nematode infection. *Microbes Infect*. 2005;7:990-996.
33. Chehayeb JF, Robertson AP, Martin RJ, Geary TG. Proteomic analysis of adult *Ascaris suum* fluid compartments and secretory products. *PLoS Negl Trop Dis*. 2014;8:e2939.
34. Ahumada V, García E, Dennis R, et al. IgE responses to *Ascaris* and mite tropomyosins are risk factors for asthma. *Clin Exp Allergy*. 2015;45:1189-1200.
35. Xia Y, Spence HJ, Moore J, et al. The ABA-1 allergen of *Ascaris lumbricoides*: sequence polymorphism, stage and tissue-specific expression, lipid binding function, and protein biophysical properties. *Parasitology*. 2000;120(Pt 2):211-224.
36. Kennedy MW. The polyprotein allergens of nematodes (NPAs) - structure at last, but still mysterious. *Exp Parasitol*. 2011;129:81-84.
37. Santos AB, Rocha GM, Oliver C, et al. Cross-reactive IgE antibody responses to tropomyosins from *Ascaris lumbricoides* and cockroach. *J Allergy Clin Immunol*. 2008;121(1040-1046):e1041.
38. Acevedo N, Erler A, Briza P, et al. Allergenicity of *Ascaris lumbricoides* tropomyosin and IgE sensitization among asthmatic patients in a tropical environment. *Int Arch Allergy Immunol*. 2011;154:195-206.
39. Ayuso R, Reese G, Leong-Kee S, Plante M, Lehrer SB. Molecular basis of arthropod cross-reactivity: IgE-binding cross-reactive epitopes of shrimp, house dust mite and cockroach tropomyosins. *Int Arch Allergy Immunol*. 2002;129:38-48.
40. Acevedo N, Mohr J, Zakzuk J, et al. Proteomic and immunochemical characterization of glutathione transferase as a new allergen of the nematode *Ascaris lumbricoides*. *PLoS ONE*. 2013;8:e78353.
41. Zakzuk J, Bornacelly A, Mercado D, et al. The evolution of IgE sensitization to *Ascaris* allergenic components in early infancy; 2014:29-31.
42. Mueller GA, Pedersen LC, Glesner J, et al. Analysis of glutathione S-transferase allergen cross-reactivity in a North American population: relevance for molecular diagnosis. *J Allergy Clin Immunol*. 2015;136:1369-1377.
43. Santiago HC, LeeVan E, Bennuru S, et al. Molecular mimicry between cockroach and helminth glutathione S-transferases promotes cross-reactivity and cross-sensitization. *J Allergy Clin Immunol*. 2012;130(248-256):e249.
44. Lozano AKY, Lopez J, Bustillo J, Caraballo L, Zakzuk J. Description of a new allergenic member of the glutathione transferase (GST) family from *Ascaris* with Omega-class features. *J Allergy Clin Immunol*. 2018;141:AB176.
45. Fernandez-Caldas E, Puerta L, Mercado D, Lockey RF, Caraballo LR. Mite fauna, Der p I, Der f I and *Blomia tropicalis* allergen levels in a tropical environment. *Clin Exp Allergy*. 1993;23:292-297.
46. Fernandez-Caldas E, Puerta L, Caraballo L, Mercado D, Lockey RF. Sequential determinations of *Dermatophagoides* spp. allergens in a tropical city. *J Investig Allergol Clin Immunol*. 1996;6:98-102.
47. Puerta L, Fernandez-Caldas E, Mercado D, Lockey RF, Caraballo LR. Sequential determinations of *Blomia tropicalis* allergens in mattress and floor dust samples in a tropical city. *J Allergy Clin Immunol*. 1996;97:689-691.
48. Caraballo L, Acevedo N. Allergy in the tropics: the impact of cross-reactivity between mites and ascaris. *Front Biosci (Elite Ed)*. 2011;3:51-64.
49. Vergara C, Caraballo L, Mercado D, et al. African ancestry is associated with risk of asthma and high total serum IgE in a population from the Caribbean Coast of Colombia. *Hum Genet*. 2009;125:565-579.
50. Vergara C, Jiménez S, Acevedo N, et al. Association of G-protein-coupled receptor 154 with asthma and total IgE in a population of the Caribbean coast of Colombia. *Clin Exp Allergy*. 2009;39:1558-1568.
51. Vergara C, Murray T, Rafaels N, et al. African ancestry is a risk factor for asthma and high total IgE levels in African admixed populations. *Genet Epidemiol*. 2013;37:393-401.
52. Haahtela T, Holgate S, Pawankar R, et al. The biodiversity hypothesis and allergic disease: world allergy organization position statement. *World Allergy Organ J*. 2013;6:3.
53. Caraballo L. The tropics, helminth infections and hygiene hypotheses. *Expert Rev Clin Immunol*. 2018;14:99-102.
54. Ardura-García C, Vaca M, Oviedo G, et al. Risk factors for acute asthma in tropical America: a case-control study in the City of Esmeraldas, Ecuador. *Pediatr Allergy Immunol*. 2015;26:423-430.
55. Strina A, Barreto ML, Cooper PJ, Rodrigues LC. Risk factors for non-atopic asthma/wheeze in children and adolescents: a systematic review. *Emerg Themes Epidemiol*. 2014;11:5.
56. Platts-Mills TA. The allergy epidemics: 1870-2010. *J Allergy Clin Immunol*. 2015;136:3-13.
57. Gelber LE, Seltzer LH, Bouzoukis JK, et al. Sensitization and exposure to indoor allergens as risk factors for asthma among patients presenting to hospital. *Am Rev Respir Dis*. 1993;147:573-578.
58. Caraballo L, Puerta L, Fernandez-Caldas E, Lockey RF, Martínez B. Sensitization to mite allergens and acute asthma in a tropical environment. *J Investig Allergol Clin Immunol*. 1998;8:281-284.
59. Andiappan AK, Puan KJ, Lee B, et al. Allergic airway diseases in a tropical urban environment are driven by dominant mono-specific sensitization against house dust mites. *Allergy*. 2014;69:501-509.
60. Richgels PK, Yamani A, Chougnet CA, Lewkowich IP. Maternal house dust mite exposure during pregnancy enhances severity of house dust mite-induced asthma in murine offspring. *J Allergy Clin Immunol*. 2017;140(1404-1415):e1409.
61. Marks GB, Zhou J, Yang HS, et al. Cord blood mononuclear cell cytokine responses in relation to maternal house dust mite allergen exposure. *Clin Exp Allergy*. 2002;32:355-360.
62. Sanchez-Borges M, Fernandez-Caldas E, Thomas WR, et al. International consensus (ICON) on: clinical consequences of mite hypersensitivity, a global problem. *World Allergy Organ J*. 2017;10:14.
63. Lynch NR, Di Prisco-Fuenmayor MC, Soto JM. Diagnosis of atopic conditions in the tropics. *Ann Allergy*. 1983;51:547-551.
64. Lynch NR, Di Prisco-Fuenmayor MC. High allergic reactivity in a tropical environment. *Clin Allergy*. 1984;14:233-240.
65. Hunninghake GM, Soto-Quiros ME, Avila L, et al. Sensitization to *Ascaris lumbricoides* and severity of childhood asthma in Costa Rica. *J Allergy Clin Immunol*. 2007;119:654-661.
66. Joubert JR, van Schalkwyk DJ, Turner KJ. *Ascaris lumbricoides* and the human immunogenic response: enhanced IgE-mediated reactivity to common inhaled allergens. *S Afr Med J*. 1980;57:409-412.
67. Lee TD, Xie CY. IgE regulation by nematodes: the body fluid of *Ascaris* contains a B-cell mitogen. *J Allergy Clin Immunol*. 1995;95:1246-1254.
68. Caraballo L, Acevedo N. New allergens of relevance in tropical regions: the impact of *Ascaris lumbricoides* infections. *WAO J*. 2011;4:77-84.

69. Caraballo LAN, Buendía E. Human Ascariasis increases the allergic response and allergic symptoms. *Curr Trop Med Rep*. 2015;2:224-232.
70. Rodriguez A, Vaca MG, Chico ME, et al. Rural to urban migration is associated with increased prevalence of childhood wheeze in a Latin-American city. *BMJ Open Respir Res*. 2017;4:e000205.
71. Britton J. Parasites, allergy, and asthma. *Am J Respir Crit Care Med*. 2003;168:266-267.
72. Pinelli E, Willers SM, Hoek D, et al. Prevalence of antibodies against *Ascaris suum* and its association with allergic manifestations in 4-year-old children in The Netherlands: the PIAMA birth cohort study. *Eur J Clin Microbiol Infect Dis*. 2009;28:1327-1334.
73. Santiago Hda C, Ribeiro-Gomes FL, Bennuru S, Nutman TB. Helminth infection alters IgE responses to allergens structurally related to parasite proteins. *J Immunol*. 2015;194:93-100.
74. Suzuki M, Hara M, Ichikawa S, et al. Presensitization to *Ascaris* antigens promotes induction of mite-specific IgE upon mite antigen inhalation in mice. *Allergol Int*. 2016;65:44-51.
75. Hagel I, Cabrera M, Hurtado MA, et al. Infection by *Ascaris lumbricoides* and bronchial hyper reactivity: an outstanding association in Venezuelan school children from endemic areas. *Acta Trop*. 2007;103:231-241.
76. Lopez N, de Barros-Mazon S, Vilela MM, Condino Neto A, Ribeiro JD. Are immunoglobulin E levels associated with early wheezing? A prospective study in Brazilian infants. *Eur Respir J*. 2002;20:640-645.
77. Moncayo AL, Vaca M, Oviedo G, et al. Effects of geohelminth infection and age on the associations between allergen-specific IgE, skin test reactivity and wheeze: a case-control study. *Clin Exp Allergy*. 2013;43:60-72.
78. Endara P, Vaca M, Platts-Mills TA, et al. Effect of urban vs. rural residence on the association between atopy and wheeze in Latin America: findings from a case-control analysis. *Clin Exp Allergy*. 2015;45:438-447.
79. Zakzuk J, Casadiego S, Mercado A, Alvis-Guzman N, Caraballo L. *Ascaris lumbricoides* infection induces both, reduction and increase of asthma symptoms in a rural community. *Acta Trop*. 2018;187:1-4.
80. Takeuchi T, Nishiyama K, Saito S, et al. Preparation of a polyclonal antibody that recognizes a unique galactose β 1-4fucose disaccharide epitope. *Carbohydr Res*. 2015;412:50-55.
81. van Ree R. Carbohydrate epitopes and their relevance for the diagnosis and treatment of allergic diseases. *Int Arch Allergy Immunol*. 2002;129:189-197.
82. Commins SP, Kelly LA, Ronmark E, et al. Galactose- α 1,3-galactose-specific IgE is associated with anaphylaxis but not asthma. *Am J Respir Crit Care Med*. 2012;185:723-730.
83. Harris NL, Loke P. Recent advances in type-2-cell-mediated immunity: insights from helminth infection. *Immunity*. 2017;47:1024-1036.
84. Itami DM, Oshiro TM, Araujo CA, et al. Modulation of murine experimental asthma by *Ascaris suum* components. *Clin Exp Allergy*. 2005;35:873-879.
85. van Riet E, Everts B, Retra K, et al. Combined TLR2 and TLR4 ligation in the context of bacterial or helminth extracts in human monocyte derived dendritic cells: molecular correlates for Th1/Th2 polarization. *BMC Immunol*. 2009;10:9.
86. Hamid F, Versteeg SA, Wiria AE, et al. Molecular diagnostics and lack of clinical allergy in helminth-endemic areas in Indonesia. *J Allergy Clin Immunol*. 2017;140(1196-1199):e1196.
87. van Riet E, Wuhler M, Wahyuni S, et al. Antibody responses to *Ascaris*-derived proteins and glycolipids: the role of phosphorylcholine. *Parasite Immunol*. 2006;28:363-371.
88. Paterson JC, Garside P, Kennedy MW, Lawrence CE. Modulation of a heterologous immune response by the products of *Ascaris suum*. *Infect Immun*. 2002;70:6058-6067.
89. Marretta J, Casey FB. Effect of *Ascaris suum* and other adjuvants on the potentiation of the IgE response in guinea-pigs. *Immunology*. 1979;37:609-613.
90. Stromberg BE. Potentiation of the reaginic (IgE) antibody response to ovalbumin in the guinea pig with a soluble metabolic product from *Ascaris suum*. *J Immunol*. 1980;125:833-836.
91. Lee TD, McGibbon A. Potentiation of IgE responses to third-party antigens mediated by *Ascaris suum* soluble products. *Int Arch Allergy Immunol*. 1993;102:185-190.
92. Zakzuk J, Bornacelly A, Mercado D, Sánchez J, Acevedo N, Caraballo L. The evolution of IgE sensitization to *Ascaris* allergenic components in early infancy. In: Galli SJ, Kim Y-Y, eds. *Allergic diseases: from mechanisms to cure*. Pisa, Italy: Pacini Editore; 2014:29-31.
93. Matricardi PM, Kleine-Tebbe J, Hoffmann HJ, et al. EAACI molecular allergology user's guide. *Pediatr Allergy Immunol*. 2016;27(Suppl 23):1-250.
94. Cantillo JF, Puerta L, Lafosse-Marin S, et al. Identification and characterization of IgE-binding tropomyosins in *Aedes aegypti*. *Int Arch Allergy Immunol*. 2016;170:46-56.
95. Cantillo JF, Puerta L, Lafosse-Marin S, et al. Allergens involved in the cross-reactivity of *Aedes aegypti* with other arthropods. *Ann Allergy Asthma Immunol*. 2017;118:710-718.
96. Carvalho Kdos A, de Melo-Neto OP, Magalhaes FB, et al. *Blomia tropicalis* Blo t 5 and Blo t 21 recombinant allergens might confer higher specificity to serodiagnostic assays than whole mite extract. *BMC Immunol*. 2013;14:11.
97. Zakzuk J, Jimenez S, Cheong N, et al. Immunological characterization of a Blo t 12 isoallergen: identification of immunoglobulin E epitopes. *Clin Exp Allergy*. 2009;39:608-616.
98. Boitelle A, Scales HE, Di Lorenzo C, et al. Investigating the impact of helminth products on immune responsiveness using a TCR transgenic adoptive transfer system. *J Immunol*. 2003;171:447-454.
99. Geiger SM, Massara CL, Bethony J, et al. Cellular responses and cytokine profiles in *Ascaris lumbricoides* and *Trichuris trichiura* infected patients. *Parasite Immunol*. 2002;24:499-509.
100. Figueiredo CA, Barreto ML, Rodrigues LC, et al. Chronic intestinal helminth infections are associated with immune hyporesponsiveness and induction of a regulatory network. *Infect Immun*. 2010;78:3160-3167.
101. Reina Ortiz M, Schreiber F, Benitez S, et al. Effects of chronic ascariasis and trichuriasis on cytokine production and gene expression in human blood: a cross-sectional study. *PLoS Negl Trop Dis*. 2011;5:e1157.
102. Hagel I, Cabrera M, Puccio F, et al. Co-infection with *Ascaris lumbricoides* modulates protective immune responses against *Giardia duodenalis* in school Venezuelan rural children. *Acta Trop*. 2011;117:189-195.
103. Cooper PJ, Chico ME, Vaca MG, et al. Effect of early life Geohelminth infections on the development of wheezing at 5 years of age. *Am J Respir Crit Care Med*. 2017;197(3):364-372.
104. Titz TO, de Araujo CAA, Enobe CS, et al. *Ascaris suum* infection modulates inflammation: implication of CD4(+) CD25(high) Foxp3(+) T cells and IL-10. *Parasite Immunol*. 2017;39:e12453.
105. Midttun HLE, Acevedo N, Skallerup P, et al. *Ascaris suum* infection downregulates inflammatory pathways in the pig intestine in vivo and in human dendritic cells in vitro. *J Infect Dis*. 2018;217:310-319.
106. Dowling DJ, Noone CM, Adams PN, et al. *Ascaris lumbricoides* pseudocoelomic body fluid induces a partially activated dendritic cell phenotype with Th2 promoting ability in vivo. *Int J Parasitol*. 2011;41:255-261.
107. Almeida S, Nejsun P, Williams AR. Modulation of human macrophage activity by *Ascaris* antigens is dependent on macrophage polarization state. *Immunobiology*. 2017;223(4-5):405-412.

108. Rocha FA, Leite AK, Pompeu MM, et al. Protective effect of an extract from *Ascaris suum* in experimental arthritis models. *Infect Immun*. 2008;76:2736-2745.
109. Deehan MR, Goodridge HS, Blair D, et al. Immunomodulatory properties of *Ascaris suum* glycosphingolipids - phosphorylcholine and non-phosphorylcholine-dependent effects. *Parasite Immunol*. 2002;24:463-469.
110. Kean DE, Ohtsuka I, Sato K, et al. Dissecting *Ascaris* glycosphingolipids for immunomodulatory moieties—the use of synthetic structural glycosphingolipid analogues. *Parasite Immunol*. 2006;28:69-76.
111. Antunes MF, Titz TO, Batista IF, et al. Immunosuppressive PAS-1 is an excretory/secretory protein released by larval and adult worms of the ascarid nematode *Ascaris suum*. *J Helminthol*. 2015;89:367-374.
112. Jex AR, Liu S, Li B, et al. *Ascaris suum* draft genome. *Nature*. 2011;479:529-533.
113. Coronado S, Barrios L, Zakzuk J, et al. A recombinant cystatin from *Ascaris lumbricoides* attenuates inflammation of DSS-induced colitis. *Parasite Immunol*. 2017;39:e12425.
114. Venugopal G, Mueller M, Hartmann S, Steinfelder S. Differential immunomodulation in human monocytes versus macrophages by filarial cystatin. *PLoS ONE*. 2017;12:e0188138.
115. Coronado S, Zakzuk J, Regino R, et al. Cystatin from the nematode *Ascaris lumbricoides* reduces inflammation in a mouse model of allergic asthma. *Allergy*. 2017;72:386-387.
116. Coronado SMM, Zakzuk J, Caraballo L. *Ascaris lumbricoides* cystatin induces specific IgE but not allergic response. *Front Immunol*. 2015. <https://doi.org/10.3389/conf.fimmu.2015.05.00295>.

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