

Factors associated with degree of atopy in Latino children in a nationwide pediatric sample: The Genes-environments and Admixture in Latino Asthmatics (GALA II) study

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Background: Atopy varies by ethnicity, even within Latino groups. This variation might be due to environmental, sociocultural, or genetic factors.

Objective: We sought to examine risk factors for atopy within a nationwide study of US Latino children with and without asthma.

Methods: Aeroallergen skin test responses were analyzed in 1830 US Latino subjects. Key determinants of atopy included country/region of origin, generation in the United States, acculturation, genetic ancestry, and site to which subjects migrated. Serial multivariate zero-inflated negative binomial regressions stratified by asthma status examined the association of each key determinant variable with the number of positive skin test responses. In addition, the independent effect of each key variable was determined by including all key variables in the final models. **Results:** In baseline analyses African ancestry was associated with 3 times (95% CI, 1.62-5.57) as many positive skin test responses in asthmatic participants and 3.26 times (95% CI, 1.02-

10.39) as many positive skin test responses in control participants. Generation and recruitment site were also associated with atopy in crude models. In final models adjusted for key variables, asthmatic patients of Puerto Rican ($\exp[\beta]$ [95% CI], 1.31 [1.02-1.69]) and mixed ($\exp[\beta]$ [95% CI], 1.27 [1.03-1.56]) ethnicity had a greater probability of positive skin test responses compared with Mexican asthmatic patients. Ancestry associations were abrogated by recruitment site but not region of origin. **Conclusions:** Puerto Rican ethnicity and mixed origin were associated with degree of atopy within US Latino children with asthma. African ancestry was not associated with degree of atopy after adjusting for recruitment site. Local environment variation, represented by site, was associated with degree of sensitization. (J Allergy Clin Immunol 2013;■■■■:■■■-■■■.)

Key words: Latino, atopy, region of origin, genetic ancestry, immigration, skin test, aeroallergen

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Abbreviations used

GALA II: Genes-environments and Admixture in Latino Asthmatics
 OR: Odds ratio
 SES: Socioeconomic status
 SNP: Single nucleotide polymorphism
 ZINB: Zero-inflated negative binomial

Atopy and atopic disorders, such as asthma, are thought to be caused by the interaction of genetic, social, and environmental factors.¹ Although atopy is understudied among US minority populations, there are well-described ethnic differences in atopic diseases, such as asthma. Asthma prevalence and severity differ between Latino groups, with Puerto Ricans having the highest prevalence and severity of asthma and Mexicans having among the lowest burden.^{2,3} There are many potential explanations for this observation, including place of birth and generation in the United States,⁴⁻⁷ acculturation,⁸ early-life exposures,⁹⁻¹² and genetic predisposition.^{1,13,14} Prior research has demonstrated that African ancestry is associated with asthma in populations of African origin^{15,16} and with IgE levels in Afro-Caribbean populations.¹⁵

Many studies evaluating ethnicity as a risk factor for atopy do not disaggregate Latinos by country or region of origin.^{14,17-20} Studies of atopy in Latino populations have either been small in total numbers,^{21,22} inadequately representative of different national origins,^{23,24} or both.²¹ Furthermore, few studies have evaluated factors that are associated with atopy in US Latino populations.^{22,25} Given the size of the US Latino population²⁶ and the diversity of this group regarding country of origin, it is important to describe patterns of atopy and determine factors contributing to variable childhood atopy susceptibility in this population.

Latinos are admixed and share varying proportions of African, Native American, and European ancestry.^{27,28} The complexity of genetic ancestry among Latinos can complicate biomedical research studies in this population. On the other hand, precisely because of this complexity, Latinos also present a unique opportunity to disentangle the clinical, social, environmental, and genetic underpinnings of population differences in health outcomes. Specifically, the mixed ancestry of Latinos provides the intrinsic variability needed to untangle complex gene-environment interactions, which might help to explain the striking differences in atopic disorders seen among Latinos and other populations.

We sought to determine whether country of origin, genetic ancestry, generation, and acculturation were associated with atopy in US Latino children. To study this question, we leveraged a large multicenter study of US Latino children with and without asthma to examine the independent effects of local environment, sociocultural factors relevant to immigration, and genetic ancestry on atopy.

METHODS**Recruitment**

Latino children were enrolled as a part of the ongoing Genes-environments and Admixture in Latino Americans (GALA II) case-control study. From July 2008 through November 2011, 4157 children (2022 asthmatic participants and 2135 healthy control subjects) were recruited from 5 centers (Chicago, Illinois; Bronx, New York; Houston, Texas; the San Francisco Bay Area, California; and Puerto Rico) by using a combination of community- and clinic-based recruitment. Participants were eligible if they were 8 to 21 years

of age and all 4 grandparents self-identified as Latino. Asthma cases were defined as participants with a history of physician-diagnosed asthma and the presence of 2 or more symptoms of coughing, wheezing, or shortness of breath in the 2 years preceding enrollment. Healthy control subjects were recruited from the community and clinics with the same catchment area as cases. Control subjects were defined as participants with no reported history of asthma, lung disease, or chronic illness over their lifetime and no reported symptoms of coughing, wheezing, or shortness of breath in the last 2 years. Control subjects were frequency matched on age (within 1 year), sex, and study center. Participants were excluded if they reported any of the following: (1) 10 or more pack years of smoking; (2) any smoking within 1 year of recruitment date; (3) history of lung diseases other than asthma (cases) or chronic illness (cases and control subjects); or (4) pregnancy in the third trimester. All local institutional review boards approved the study, and all participants/parents provided appropriate written assent/consent.

Trained interviewers, proficient in both English and Spanish, administered questionnaires to gather baseline demographic data, as well as information on general health, asthma status, acculturation, social, and environmental exposures. For this analysis, only mainland US participants who had skin prick test data available were studied. Puerto Rican subjects from Puerto Rico were not included because the analysis is focused on patterns of atopy in the mainland United States and evaluates factors, including acculturation and migrant generation, that are not relevant to this population. This resulted in 1129 participants with asthma and 1167 healthy control subjects. Participants missing data on age ($n = 8$), mother's education ($n = 31$), first-born child status ($n = 36$), breast-feeding ($n = 83$), mother smoking during pregnancy ($n = 12$), postnatal smoking exposure ($n = 249$), cat or dog ownership in the first year of life ($n = 11$), and day care attendance in the first 3 years of life ($n = 24$) were excluded from the analyses, yielding an analytic sample size of 1830 (914 asthmatic patients and 916 control subjects).

Outcome measurements

Skin testing was performed on the volar aspect of the forearm with the Multi-Test II Device (Lincoln Diagnostics, Decatur, Ill) for a panel of 14 aeroallergens, including dust mite (*Dermatophagoides pteronyssinus* and *Dermatophagoides farinae*), dog, cat, cockroach (mix of German and American cockroach), mouse epithelium, rat epithelium, tree (oak and olive/elm), grass (perennial ryegrass and timothy grass), short ragweed, *Alternaria tenuis*, *Hor-modendrum cladosporioides*, and *Aspergillus* mix. Any allergen eliciting a mean wheal diameter of at least 3 mm greater than that elicited by the saline control was considered positive. The number of positive skin test responses was summed for each subject to provide the count variable used in these analyses. For secondary analyses, we classified any subject with a positive test response as atopic and those with all negative test responses as nonatopic.

Key independent variables

Origin. The participant's country of birth, parents' country of birth, and self-reported country of birth of all 4 grandparents were used to determine country or region of origin. First, for participants born outside the United States, their origin was determined by their country of birth. For US-born participants, origin was determined by their parents' country of birth (or grandparents' country of origin if their parents were born in the United States). For those with missing information for 1 parent, origin defaulted to the origin of the known parent. Participants were then classified as Puerto Rican, Mexican, South American, Central American, non-Puerto Rican Caribbean, or mixed Latino (for subjects of multiple origins who did not fit in the prior mentioned categories).

Generation in the United States. The same variables used to assign the participant's origin were also used to determine their generation of immigration. Foreign-born participants were classified as first-generation immigrants, US-born participants with at least 1 foreign-born parent were classified as second-generation immigrants, and US-born participants with both US-born parents were classified as third-generation immigrants or greater. For participants with missing information for 1 parent, immigrant status was determined from information of the known parent.

TABLE I. Characteristics of GALA II case-control study participants: 2006-2011

	Total no. of subjects (%)	Cases, no. (%)	Control subjects, no. (%)	P value*
No.	1830 (100)	914 (100)	916 (100)	
Total no. of positive skin prick test responses†	2 (0.4)	3 (1.5)	1 (0.3)	<.001
Demographics				
Average age‡	13.1 (3.4)	12.5 (3.2)	13.6 (3.5)	<.001
Male sex	894 (48.9)	504 (55.1)	390 (42.6)	<.001
Region of origin				<.001
Mexico	1057 (57.8)	491 (53.7)	566 (61.8)	
Puerto Rico	223 (12.2)	152 (16.6)	71 (7.8)	
Central America	190 (10.4)	85 (9.3)	105 (11.5)	
Caribbean	95 (5.2)	36 (3.9)	59 (6.4)	
South America	33 (1.8)	11 (1.2)	22 (2.4)	
Mixed	226 (12.3)	134 (14.7)	92 (10)	
Missing data	6 (0.3)	5 (0.5)	1 (0.1)	
Global ancestry				
Average % of African ancestry‡	10.3 (12.3)	11.2 (12.7)	9.3 (11.7)	<.001
Average % of European ancestry‡	43.4 (18.4)	46.2 (17.6)	40.5 (18.9)	<.001
Average % of Native American ancestry‡	46.3 (25.2)	42.5 (24.1)	50.2 (25.9)	<.001
Missing data	183 (10)	71 (7.8)	112 (12.2)	
Generation in the US				
First generation	338 (18.5)	92 (10.1)	246 (26.9)	<.001
Second generation	1285 (70.2)	687 (75.2)	598 (65.3)	
Third generation or later	178 (9.7)	121 (13.2)	57 (6.2)	
Missing data/not applicable	29 (1.6)	14 (1.5)	15 (1.6)	
Mother's acculturation level				
Foreign born, living in US <10 y, prefers Spanish	241 (13.2)	73 (8)	168 (18.3)	<.001
Foreign born, living in US 10-15 y, prefers Spanish	357 (19.5)	177 (19.4)	180 (19.7)	
Foreign born, living in US ≥15 y, prefers Spanish	706 (38.6)	341 (37.3)	365 (39.8)	
Foreign born, living in US ≥10 y, prefers English	116 (6.3)	70 (7.7)	46 (5)	
US born, prefers English	314 (17.2)	205 (22.4)	109 (11.9)	
Missing data/not applicable	96 (5.2)	48 (5.3)	48 (5.2)	
Mother's education level				
Bachelor's degree and greater	89 (4.9)	47 (5.1)	42 (4.6)	.561
High school degree	782 (42.7)	399 (43.7)	383 (41.8)	
No high school degree	959 (52.4)	468 (51.2)	491 (53.6)	
Variables associated with sensitization				
First-born child	875 (47.8)	398 (43.5)	477 (52.1)	<.001
Breast-fed	1316 (71.9)	605 (66.2)	711 (77.6)	<.001
Mother smoking during pregnancy	89 (4.9)	54 (5.9)	35 (3.8)	.049
Adults smoking during pregnancy	588 (32.1)	329 (36.0)	259 (28.3)	<.001
Owned dog/cat in first year of life	511 (27.9)	235 (25.7)	276 (30.1)	.040
Attended day care	427 (23.3)	242 (26.5)	185 (20.2)	.002

Maternal acculturation. Acculturation status was determined by using the mother's characteristics as a proxy for the child's home environment because early-life factors⁹⁻¹² have been shown to be important atopy risk factors, and it is not known whether these factors would differ depending on the degree of US acculturation. Consistent with previous studies,²⁹⁻³² acculturation status was determined by aggregating the reported country of birth for the mother, the language preference of the mother, and the length of time the mother had been living in the United States. A 5-level acculturation variable was constructed: US-born mother who prefers English, foreign-born mother living in the United States for 10 or more years and prefers English, foreign-born mother living in the United States for 15 or more years and prefers Spanish, foreign-born mother living in the United States for 10 to 15 years and prefers Spanish, and foreign-born mother living in the United States for less than 10 years and prefers Spanish.

Ancestry proportions. To estimate the ancestral admixture of GALA II participants, we used available genome-wide genotyping data from the Affymetrix Axiom Latino Array (Affymetrix, Santa Clara, Calif).³³ We removed single nucleotide polymorphisms (SNPs) with greater than 5% missing values and failing platform-specific SNP quality criteria ($n = 63,328$), along with those out of Hardy-Weinberg equilibrium ($n = 1845$, $P < 10^{-6}$) within their respective populations. After combining HapMap European (CEU) and African (YRI) ancestral data with Native American samples (kindly provided by Cheryl Winkler, Andres Moreno, Karla Sandoval, and Carlos Bustamante), a final sample of 568,037 autosomal SNPs with relevant ancestral data were used to estimate the 3-way ancestry using the program ADMIXTURE.³⁴ Native American and African ancestry were chosen for modeling because African and European ancestries are along the same axis of assortment in Latino populations.

Recruitment site/site to which migration occurred. We included recruitment site as a proxy for unmeasured local environmental factors, which can vary by site.

Covariates. Covariates for all models were selected based on the literature and included sex,³⁵ age,^{36,37} mother's highest education level, first-born child status,³⁸ breast-feeding,³⁹ mother smoking during pregnancy,⁴⁰ postnatal smoking exposure,⁴¹ cat or dog ownership in the first year of life,⁴²⁻⁴⁴ and day care attendance in the first 3 years of life.^{45,46} Aside from *a priori* covariates, a number of regional air pollution exposure variables (particulate matter of 2.5-10 μm in diameter, particulate matter <2.5 μm in diameter, ozone, and distance from major traffic arteries) were evaluated. Estimates for each subject were based on geocoded addresses at 1 to 3 years of age and at the time of recruitment mapped to the US Environmental Protection Agency's Air Quality System Data and averaged for each year. These were subsequently removed from the analyses because of lack of significance.

Statistical methods

Descriptive statistics for selected variables were presented for the overall study population and according to asthma status. All analyses were stratified by asthma status to avoid inflating the effects of asthma.⁴⁷ To determine significant differences between children with and without asthma, we used the Mann-Whitney test, t tests, and χ^2 tests. Zero-inflated negative binomial (ZINB) regression models were used to estimate associations of each key variable (origin, recruitment site, ancestry proportions, generation, and acculturation) with the number of positive skin test responses. ZINB regression models count data with an excess of zeros and assumes the zeros are distributed from a different process than nonzero observations (the "count portion") and are thus modeled independently. The count portion models the negative binomial distribution (log link), and the mass at zero models a binary distribution. Because we are interested in the contribution of the relevant exposures to the number of positive skin test responses to aeroallergens, we extract the coefficients from

*P values were calculated from the Mann-Whitney test (all ordinal variables), t tests (all continuous variables), and χ^2 tests (all categorical nonranked variables).

†Reported as median (interquartile range).

‡Reported as mean (SD).

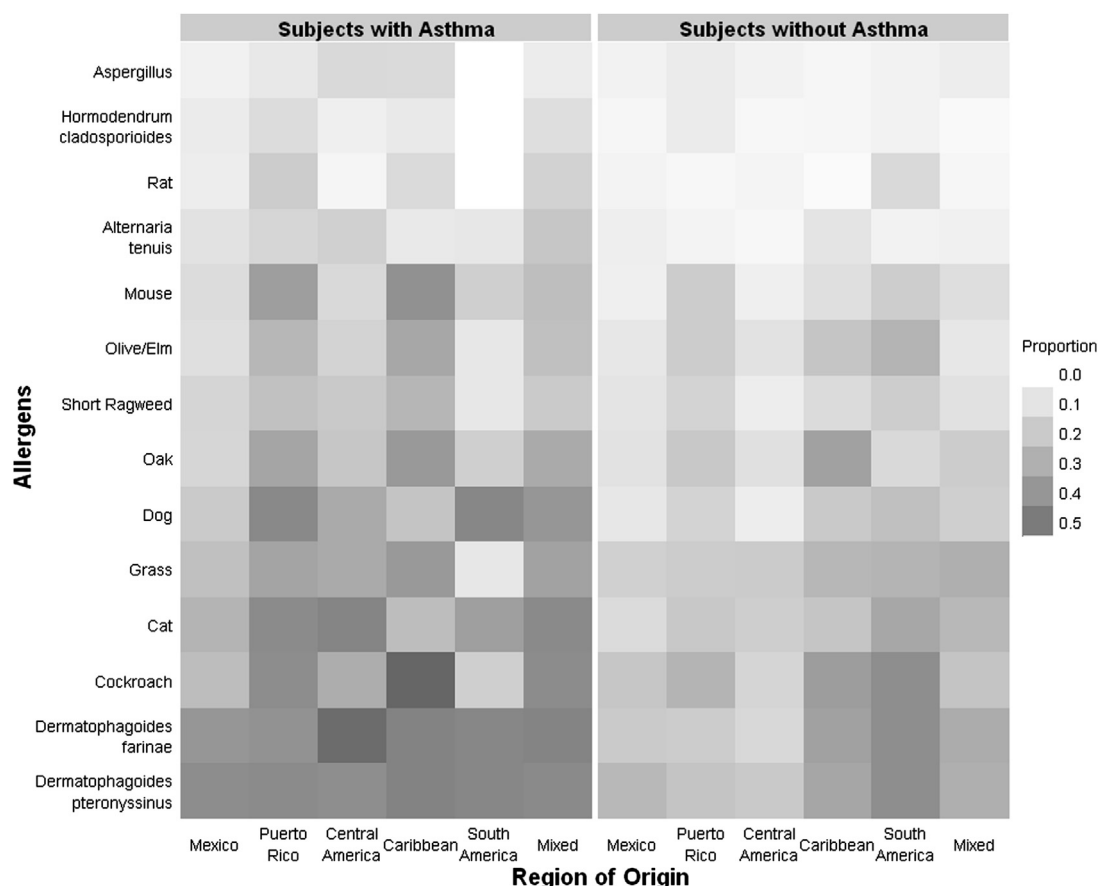


FIG 1. Proportion of participants with a positive skin test response to allergens tested by region of origin. The proportions are represented on a scale in increments of 0.1, ranging from 0 (white) to 1.0 (black).

the “count” portion of the *zeroInfl()* function in the R package (pscl), and exponentiate these to determine the expected effect of our predictors on the number of positive skin prick test responses. In addition, the independent effect of each key variable was determined by adjusting key variables in the final models stratified by children with and without asthma. Because acculturation and migrant generation were defined by using the mother’s place of birth, they were not included in the same model to avoid multicollinearity. Similarly, we created 2 separate final models for country/region of origin and recruitment site because of the collinearity of these 2 variables. As a secondary analysis, we evaluated the association of these factors with the presence of any sensitization with logistic models by using the same baseline covariates as with the models for number of positive skin test responses. These models comprised all key variables, including generation, region of origin, and ancestry proportion. All analyses were performed with the statistical software R, version 2.13.1.

RESULTS

Cohort characteristics

The characteristics of the cohort are displayed in Table I. The average age of participants was 13.1 ± 3.4 years. The 2 largest groups of participants were from Mexico (57.8%) and Puerto Rico (12.2%), with smaller numbers from South America, Central America, and the Caribbean. More than two thirds of GALA II children were second-generation migrants (70.2%) with low levels of acculturation, and 71.3% of mothers preferred Spanish. GALA II is a primarily low socioeconomic status (SES) cohort where 95.1% of participants had a maternal education level of high school or less. We also present the distribution of skin test

response positivity to each of the evaluated allergens by region of origin in Table E1 in this article’s Online Repository at www.jacionline.org. Notably, mouse and cockroach were more common sensitizers in New York.

Finally, there were differences in total number of positive skin test responses between asthmatic patients (median, 3; interquartile range, 1-5) and control subjects (median, 1; interquartile range, 0-3), respectively ($P < .001$), and any positive skin test response (77% vs 54.4% in cases and control subjects, respectively) and the sensitivity to dust mites (52.5% and 29.7% in cases and controls subjects, respectively). Along with these sensitivity differences, asthmatic participants were also different from control subjects in sex, region of origin, estimates of global ancestry, generation of migrant, acculturation, and variables associated with sensitization, including first-born child status and breast-feeding ($P < .01$).

Origin, migrant generation, ancestry, acculturation, recruitment site, and atopy

The distribution of positive skin test responses by region of origin is presented in Fig 1 for asthmatic participants and healthy control subjects, where darker squares represent a greater proportion of sensitized subjects. There were differences in the proportion of subjects sensitized to these aeroallergens by asthma status and by region of origin among both the asthmatic and nonasthmatic participants.

TABLE II. ZINB regression analysis evaluating potential primary determinants of number of positive skin prick test responses among Latino children with asthma

	Asthmatic participants		Nonasthmatic participants	
	No.	Relative no. of positive skin prick test responses (95% CI)*	No.	Relative no. of positive skin prick test responses (95% CI)*
Model of country of origin	909		915	
Mexico	491	1.0 (referent)	566	1.0 (referent)
Puerto Rico	152	1.48 (1.25-1.75)	71	1.27 (0.91-1.77)
Central America	85	1.12 (0.91-1.36)	105	1.01 (0.75-1.36)
Caribbean	36	1.39 (1.06-1.83)	59	1.34 (0.98-1.83)
South America	11	1.17 (0.64-2.15)	22	1.49 (0.95-2.35)
Mixed	134	1.40 (1.19-1.65)	92	0.97 (0.73-1.3)
Model of ancestry	843		804	
African ancestry proportion		3.00 (1.62-5.57)		3.26 (1.02-10.39)
Native American ancestry proportion		0.92 (0.64-1.31)		1.28 (0.76-2.15)
Model of generation	900		901	
First-generation migrant	92	1.0 (referent)	246	1.0 (referent)
Second-generation migrant	687	1.01 (0.81-1.26)	598	1.18 (0.94-1.5)
Third-generation migrant or later	121	1.31 (1.00-1.72)	57	1.52 (1.04-2.23)
Model of mother's acculturation	866		868	
Foreign born, in US <10 y, prefers Spanish	73	1.0 (referent)	168	1.0 (referent)
Foreign born, in US 10-15 y, prefers Spanish	177	1.1 (0.84-1.45)	180	0.97 (0.73-1.3)
Foreign born, in US ≥15 y, prefers Spanish	341	1.07 (0.83-1.38)	365	1.18 (0.9-1.54)
Foreign born, in US ≥10 y, prefers English	70	1.31 (0.95-1.8)	46	1.04 (0.66-1.64)
US born, prefers English	205	1.29 (0.99-1.69)	109	1.16 (0.83-1.63)
Model of recruitment site	914		916	
New York, New York	273	1.0 (referent)	246	1.0 (referent)
Chicago, Illinois	211	0.65 (0.56-0.76)	232	0.59 (0.47-0.74)
San Francisco Bay Area, California	300	0.51 (0.44-0.59)	341	0.72 (0.58-0.88)
Houston, Texas	130	0.69 (0.58-0.82)	97	0.51 (0.36-0.71)

Boldface type indicates significant antilog-β at an α value of .05.

*All models of key determinants are adjusted for sex, age, mother's highest education level, first-born child status, breast-feeding, prenatal smoke exposure, pet ownership in the first year of life, and day care attendance.

We display the ZINB regression-based associations between our key determinants and atopy for children with and without asthma (Table II). The associations were adjusted for the covariates as noted, but each predictor variable (region of origin, ancestry, generational status, recruitment site, and acculturation) was tested without accounting for the other predictors in this first analytic step.

TABLE III. Evaluation of recruitment site, ancestry, and migrant generation as determinants of positive skin prick test response differences among Latino children with and without asthma

	Asthmatic participants		Nonasthmatic participants	
	No.	Relative no. of positive skin prick test responses (95% CI)*	No.	Relative no. of positive skin prick test responses (95% CI)*
Model for no. of positive skin prick test responses	829		790	
New York, NY	246	1.0 (referent)	223	1.0 (referent)
Chicago, Illinois	190	0.68 (0.56-0.81)	207	0.56 (0.43-0.72)
San Francisco Bay Area, California	267	0.53 (0.44-0.64)	266	0.74 (0.58-0.95)
Houston, Texas	126	0.74 (0.6-0.9)	94	0.49 (0.35-0.7)
African ancestry proportion		1.53 (0.82-2.86)		0.8 (0.22-2.93)
Native American ancestry proportion		1.18 (0.83-1.69)		1 (0.58-1.72)
First-generation migrant		1.0 (referent)		1.0 (referent)
Second-generation migrant		0.97 (0.78-1.2)		1.19 (0.94-1.51)
Third-generation migrant or later		1.13 (0.85-1.49)		1.41 (0.96-2.08)

Boldface type indicates significant antilog-β at an α value of .05.

*Adjusted for sex, age, mother's highest education level, first-born child status, breast-feeding, prenatal smoke exposure, pet ownership in the first year of life, and day care attendance.

Among asthmatic participants, children from Puerto Rico (1.48; 95% CI, 1.25-1.75) and the Caribbean (1.39; 95% CI, 1.06-1.83) and those of mixed national origin (1.40; 95% CI, 1.19-1.65) were 40% to 50% more likely to have positive skin prick test responses (Table II) than children from Mexico. Increasing African Ancestry was also associated with 3 times as many positive skin test responses in asthmatic participants (exp[β], 3.00; 95% CI, 1.62-5.57). The association of the number of positive skin test responses with third-generation status alone (exp[β], 1.31; 95% CI, 1.00-1.72) and highest level of acculturation (exp[β], 1.29; 95% CI, 0.99-1.69) were marginally significant, but additional associations with acculturation lacked precision.

In participants without asthma, African ancestry was associated with 3.26 times as many positive skin test responses (exp[β], 3.26; 95% CI, 1.02-10.39). Native American ancestry was not associated with number of positive skin test responses. Compared with first-generation subjects, third-generation status was associated with 1.52 times as many positive skin test responses (exp[β], 1.52; 95% CI, 1.04-2.23; Table II). The association with Caribbean origin (exp[β], 1.34; 95% CI, 0.98-1.83) was marginally significant, but no other obvious national origin differences were present in control subjects.

Compared with New York City, being recruited in Chicago, the San Francisco Bay Area, or Houston was associated with lower risk of sensitization for both participants with and those without asthma. These associations remained nearly identical after controlling for ancestry and generation in the United States (Table III).

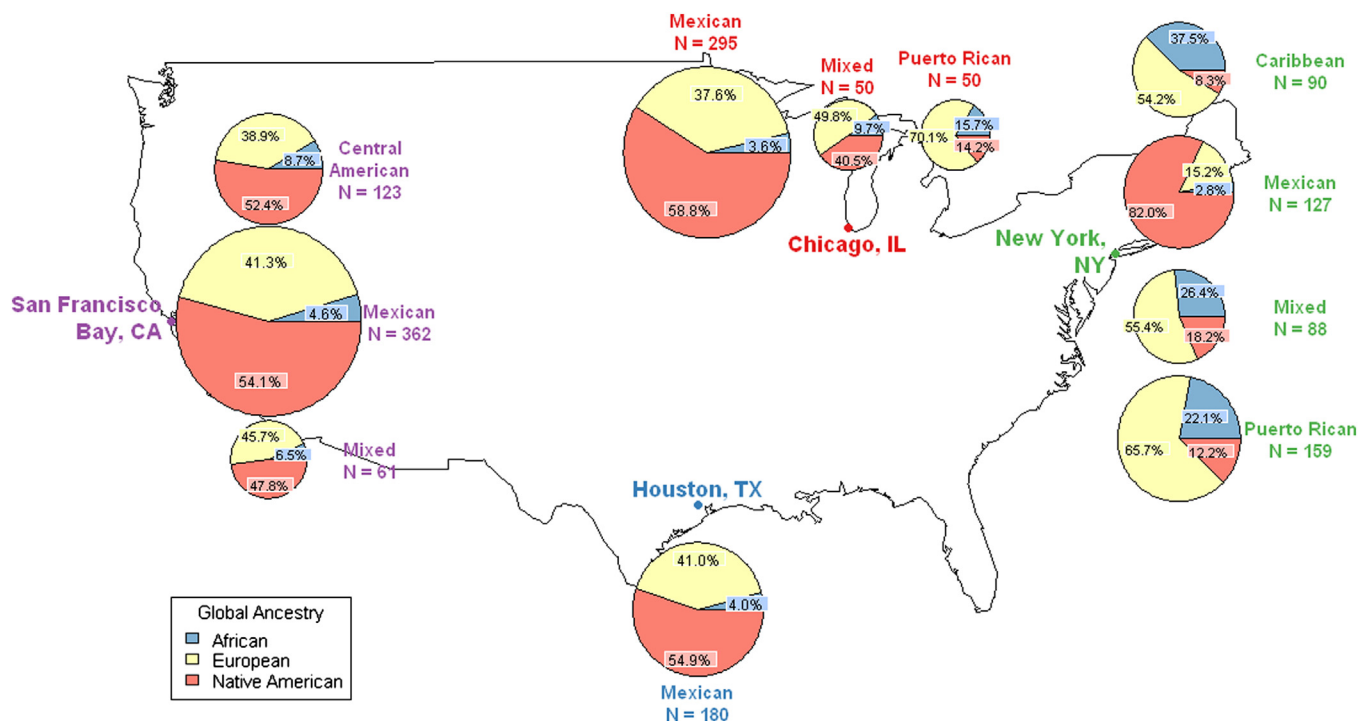


FIG 2. Global ancestry proportions of GALA II Latinos by origin and recruitment site. Diameter of pie charts represents the size of group recruited at each site. Blue, yellow, and orange wedges represent African, European, and Native American ancestry, respectively. Global ancestry proportions for population sizes of less than 20 are not displayed on this map.

Ancestry, origin, and generation of immigration

In Fig 2 we display the mean proportions of African, European, and Native American ancestry for each region of origin at each recruitment site. Participants with Caribbean or Puerto Rican origin had higher levels of African ancestry ($P < .001$) than participants with other regions of origin, which is consistent with previous studies.⁴⁸

Table III evaluated whether addition of local effects (recruitment site/site to which migration occurred) and generation of immigration status attenuated the effects of genetic ancestry. Indeed, only recruitment site remained significantly associated with atopy, abrogating the effects of African ancestry and generation of immigrant. To further examine the effect of recruitment site on ancestry associations, we evaluated whether there were differences in ancestry distribution across sites (Fig 3) and found that there were clear differences in African ancestry between New York and other sites.

Finally, we sought to investigate whether the effects of recruitment site on ancestry associations were due to differential migration patterns of Latino national origin groups. We tested whether addition of country of origin and generation of immigration status attenuated the effects of genetic ancestry (ie, whether recruitment site/local effects were proxies for migration patterns). Among participants with asthma (Table IV), the associations for African ancestry ($\exp[\beta]$, 3.13; 95% CI, 1.63-6.01) were slightly higher than the effects seen in Table II. In participants without asthma (Table IV), the magnitude of the association between African ancestry and positive skin test responses was even greater after controlling for other key variables ($\exp[\beta]$, 4.99; 95% CI, 1.19-20.91). There were no significant associations between Native American ancestry and positive skin test

responses in participants without asthma. However, there was a linear trend between generation of immigrant with the number of positive skin test responses, with third-generation Latinos having a 65% greater likelihood of positive number of skin test responses compared with first-generation Latinos ($\exp[\beta]$, 1.65; 95% CI, 1.05-2.59). These results suggest that the local effects of recruitment site are not simply proxies for differential migration patterns of different ethnic groups because region or country of origin did not abrogate the associations in the same manner as recruitment site.

As a secondary analysis, we evaluated the contribution of the key variables to the presence of any sensitization in a model that accounted for origin, ancestry, and migrant status. In asthmatic patients we found that of our key variables, only region of origin was associated with atopic status (Central America: odds ratio [OR], 3.99; 95% CI, 1.71-9.3; Caribbean: OR, 3.57; 95% CI, 1.03-12.42; and mixed: OR, 2.14; 95% CI, 1.15-3.97; reference = Mexican). Similarly, in control subjects, of our key variables, only region of origin was associated with atopic status (Puerto Rican: OR, 2.23; 95% CI, 1.01-4.92; Caribbean: OR, 3.77; 95% CI, 1.41-10.06; South American: OR, 3.37; 95% CI, 1.06-10.69; and mixed: OR, 2.54; 95% CI, 1.38-4.66; reference = Mexican).

DISCUSSION

GALA II is the largest nationwide genetic study of Latino children with and without asthma. This study has carefully collected data that allow estimation of the effects of acculturation, migration patterns, and genetic ancestry and other asthma-related factors on lung function and atopy. By leveraging these strengths of GALA II, we were able to evaluate the independent

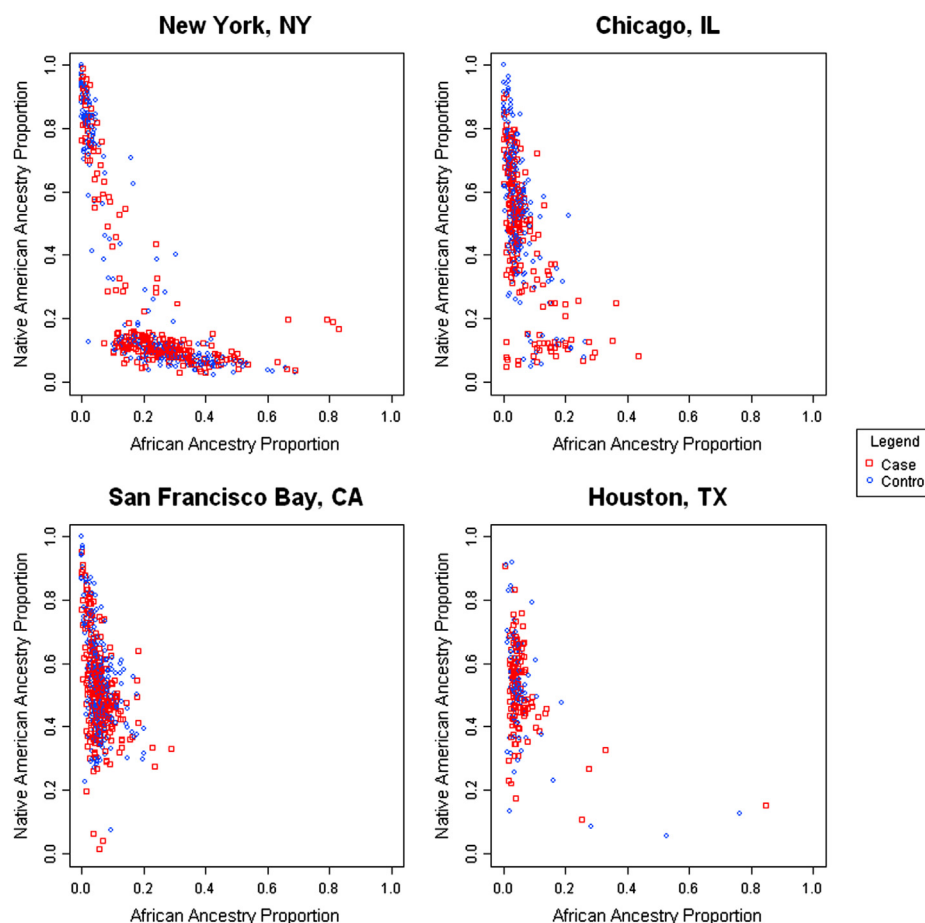


FIG 3. Ancestral proportions and recruitment site and case-control status. Each plot on these graphs represents a recruitment site. The y-axis represents global estimates of individual Native American ancestral proportion, and the x-axis represents global estimates of individual African ancestral proportion. Asthmatic patients are represented by *squares*, and nonasthmatic subjects are represented by *circles*.

associations of social, environmental, and genetic factors with degree of atopy. Our findings suggest that country/region of origin and local exposures might have the greatest influence on the degree of atopy in Latino children. In multivariable analysis genetic ancestry was not independently associated with atopy in this population. Because Latinos of different countries migrate preferentially to various sites, the associations of genetic ancestry in the United States might be proxies for local exposures.

Our findings on ancestry are in keeping with another study that examined the association of sensitization with African ancestry and did not find an ancestry-specific association with atopy in white and African American populations.⁴⁹ In that study by Yang et al,⁴⁹ the effects of ancestry on sensitization were explained by local neighborhood effects. However, our results are in contrast with the association between African ancestry and atopy reported by other investigators in Afro-Caribbean¹⁵ and multiethnic US populations.¹³ Although we did observe an association between African ancestry and degree of atopy, these results were not independent of local environmental factors. New York had the highest rates of sensitization to cockroach and mouse epithelium and also had the highest proportions of Afro-Caribbean Latino subjects. The initially observed ancestry effect might be confounded by patterns of migration to this local environment. Our results move beyond these prior investigations in that we had a large

number of children from multiple Latin American countries that migrated to a number of different regions in the United States. As such, we are uniquely positioned to differentiate genetic from socioeconomic, sociocultural, and unmeasured local factors. Environmental factors, such as SES, might covary with African ancestry¹ and atopic diseases.⁵⁰ We accounted for SES, generation status, degree of acculturation, and unmeasured local site effects that could vary by differential patterns of immigration among Latinos from different countries to different areas of the United States. Our study was also rigorous in adjusting for traditional factors affecting atopy, including sex,³⁵ age,^{36,37} mother's highest education level, first-born child status,³⁸ breast-feeding,³⁹ maternal smoking during pregnancy,⁴⁰ postnatal smoking exposure,⁴¹ cat or dog ownership in the first year of life,⁴²⁻⁴⁴ and day care attendance in the first 3 years of life.

Interestingly, differences in degree of atopy by country of origin were only seen in asthmatic participants. It is possible that some of the differential associations by asthma status are modified by other variables that covary with asthma. Although we have included a number of covariates that influence the development of atopy, it is possible that our estimates might be residually confounded by unmeasured factors that are associated with asthma and influence atopy (eg, dietary influences in early life⁹⁻¹²). Alternately, given lower levels of atopy in participants

TABLE IV. Evaluation of origin, ancestry, and migrant generation as determinants of positive skin prick test response differences among Latino children with and without asthma

	Asthmatic participants		Nonasthmatic participants	
	Relative no. of positive skin prick test responses (95% CI)*		Relative no. of positive skin prick test responses (95% CI)*	
	No.		No.	
Model for no. of positive skin prick test responses	825		790	
Region of origin				
Mexico	446	1.0 (referent)	486	1.0 (referent)
Puerto Rico	127	1.31 (1.02-1.69)	59	0.96 (0.61-1.52)
Central America	77	1 (0.81-1.23)	91	0.96 (0.69-1.32)
Caribbean	36	1.17 (0.83-1.66)	55	0.94 (0.58-1.54)
South America	11	1.03 (0.49-2.15)	19	1.55 (0.98-2.47)
Mixed	128	1.27 (1.03-1.56)	80	0.81 (0.59-1.11)
Global ancestry proportion				
African		3.13 (1.63-6.01)		4.99 (1.19-20.91)
Native American		1.47 (0.95-2.27)		1.51 (0.84-2.71)
Generation in US				
First generation		1.0 (referent)		1.0 (referent)
Second generation		0.97 (0.77-1.22)		1.21 (0.94-1.55)
Third generation		1.14 (0.84-1.55)		1.65 (1.05-2.59)

Boldface type indicates significant antilog- β at an α value of .05.

*Adjusted for sex, age, mother's highest education level, first-born child status, breast-feeding, prenatal smoke, pet ownership in the first year of life, and day care attendance.

without asthma, we might not have had sufficient power to see differences in numbers of positive skin test responses by region of origin. This last possibility is supported by the fact that we did see differences in the presence of atopy by region of origin in both asthmatic patients and control subjects.

We also found that generation status had modest and variable effects in increasing the risk of positive skin test responses to aeroallergens, which were abrogated after adjusting for recruitment site. Other studies have suggested that migration as a child (first generation) or birth in a high-income country (second generation) increases the risk of atopic disease.^{6,7,51,52} First-generation immigrants who arrive in a country as adults might have less risk of atopy than their second-generation offspring,^{6,51} suggesting that early-life exposures are important in this phenomenon. However, the majority of our first-generation participants, the reference group, were children and not first-generation adult participants. These subjects would have had similar early-life environments in the United States compared with higher-generation immigrants. Finally, we did not observe associations by acculturation status despite prior studies suggesting that US lifestyle was associated with atopy.^{53,54} One implication of this finding might be that biological factors associated with growing up in the United States modulate early-life immune development^{11,55,56} and might be more relevant than other environmental factors associated with acculturation.

Our study has a number of limitations. First, the sample size of participants from different Latin American regions varies and is smaller for participants of South American and Caribbean origin. This limits our ability to distinguish differences between groups and limits the number of Latinos with a higher proportion of African ancestry. However, the distribution by region of origin in

our sample is representative of national immigration patterns and similar to US Census proportions. Additionally, GALA II was limited to the ages of 8 to 21 years, restricting our ability to evaluate age of immigration on the development of atopic disease. Finally, there might be some residual confounding from unmeasured factors, such as discrimination and stress in pregnancy,^{57,58} that vary by skin pigmentation⁵⁹ and are also associated with atopic disease. The local effects might be due to other unmeasured environmental factors, including levels of allergen in settled dust,⁶⁰⁻⁶³ levels of endotoxin exposure,⁶⁴⁻⁶⁷ or the microbiome.⁵⁶ We examined annual regional air pollution exposures, such as particulate matter of 2.5 to 10 μm in diameter, particulate matter less than 2.5 μm in diameter, ozone, and distance from major traffic arteries, as potential covariates, but these variables were not significantly associated with atopy in this population. Finally, GALA II is a case-control design, which might affect the representativeness of our results in terms of prevalence; however, separate case-control analyses allow for interpretations independent of asthma prevalence.

Using GALA II, the largest nationwide genetic study of Latino children with and without asthma, we were able to describe patterns of atopy and also evaluate the independent contributions of factors potentially important in the US Latino population (acculturation, migration pattern, and genetic ancestry). Without this type of data, key determinants of atopy would be missed in this growing and understudied US minority group.

In summary, there are differences in atopy by country of origin among Latino participants with asthma in this nationwide sample. Furthermore, in contrast to prior studies, genetic estimates of African ancestry were not associated with increased risk of atopy in multivariate analyses and might be proxies for local environmental factors because there are differences in patterns of immigration from Latino countries to sites within the United States. Finally, our associations vary by national origin and highlight that Latinos from different countries of origin should be evaluated independently in studies of atopy.

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Clinical implications: Latino children in the United States have variable degrees of atopy based on both country of origin and local environment. In particular, Puerto Rican children are at higher risk.

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TABLE E1. Sensitization for each allergen by case-control status and recruitment region

Common allergen name	Scientific name	Chicago*	New York	San Francisco Bay Area	Texas	Total
Cases						
Total cases		211	273	300	130	914
European dust mite	<i>Dermatophagoides pteronyssinus</i>	85 (40.3)	137 (50.2)	94 (31.3)	89 (68.5)	405 (44.3)
American dust mite	<i>Dermatophagoides farinae</i>	72 (34.1)	134 (49.1)	112 (37.3)	80 (61.5)	398 (43.5)
Dog hair and dander	Dog hair and dander, mixed breeds	56 (26.5)	121 (44.3)	63 (21)	25 (19.2)	265 (29)
Cat pelt and hair	Cat pelt and hair mix	75 (35.5)	121 (44.3)	96 (32)	34 (26.2)	326 (35.7)
Cockroach	<i>Periplaneta americana</i> and <i>Blattella germanica</i>	55 (26.1)	151 (55.3)	48 (16)	49 (37.7)	303 (33.2)
Mouse epithelium	<i>Mus musculus</i>	36 (17.1)	116 (42.5)	23 (7.7)	13 (10)	188 (20.6)
Rat epithelium	<i>Rattus norvegicus</i>	20 (9.5)	57 (20.9)	12 (4)	7 (5.4)	96 (10.5)
Oak tree mix	Red, Virginia Live, and White Oak	47 (22.3)	102 (37.4)	38 (12.7)	22 (16.9)	209 (22.9)
Olive tree†	<i>Olea europaea</i>	30 (14.2)	82 (30)	31 (10.3)	17 (13.1)	160 (17.5)
Rye and timothy grass	<i>Lolium perenne</i> and <i>Phleum pratense</i>	50 (23.7)	94 (34.4)	95 (31.7)	25 (19.2)	264 (28.9)
Ragweed	<i>Ambrosia artemisiifolia</i>	40 (19)	59 (21.6)	53 (17.7)	20 (15.4)	172 (18.8)
Mold	<i>Alternaria tenuis</i>	32 (15.2)	41 (15)	31 (10.3)	25 (19.2)	129 (14.1)
Mold	<i>Hormodendrum cladosporioides</i>	21 (10)	33 (12.1)	8 (2.7)	22 (16.9)	84 (9.2)
<i>Aspergillus</i> species	<i>A. amstelodami</i> , <i>A. flavus</i> , <i>A. fumigatus</i> , <i>A. nidulans</i> , <i>A. niger</i>	12 (5.7)	25 (9.2)	17 (5.7)	13 (10)	67 (7.3)
Control subjects						
Total control subjects		232	246	341	97	916
European dust mite	<i>D. pteronyssinus</i>	61 (26.3)	87 (35.4)	71 (20.8)	30 (30.9)	249 (27.2)
American dust mite	<i>D. farinae</i>	50 (21.6)	82 (33.3)	48 (14.1)	25 (25.8)	205 (22.4)
Dog hair and dander	Dog hair and dander, mixed breeds	22 (9.5)	43 (17.5)	36 (10.6)	3 (3.1)	104 (11.4)
Cat pelt and hair	Cat pelt and hair mix	38 (16.4)	61 (24.8)	50 (14.7)	6 (6.2)	155 (16.9)
Cockroach	<i>P. americana</i> and <i>B. germanica</i>	59 (25.4)	99 (40.2)	33 (9.7)	20 (20.6)	211 (23)
Mouse epithelium	<i>M. musculus</i>	15 (6.5)	46 (18.7)	12 (3.5)	2 (2.1)	75 (8.2)
Rat epithelium	<i>R. norvegicus</i>	11 (4.7)	15 (6.1)	11 (3.2)	1 (1)	38 (4.1)
Oak tree mix	Red, Virginia Live, and White Oak	20 (8.6)	66 (26.8)	41 (12)	1 (1)	128 (14)
Olive tree†	<i>O. europaea</i>	17 (7.3)	43 (17.5)	39 (11.4)	3 (3.1)	102 (11.1)
Rye and timothy grass	<i>Lolium perenne</i> and <i>Phleum pratense</i>	30 (12.9)	72 (29.3)	70 (20.5)	9 (9.3)	181 (19.8)
Ragweed	<i>Ambrosia artemisiifolia</i>	23 (9.9)	30 (12.2)	40 (11.7)	3 (3.1)	96 (10.5)
Mold	<i>Alternaria tenuis</i>	10 (4.3)	13 (5.3)	27 (7.9)	2 (2.1)	52 (5.7)
Mold	<i>Hormodendrum cladosporioides</i>	5 (2.2)	14 (5.7)	11 (3.2)	2 (2.1)	32 (3.5)
<i>Aspergillus</i> species	<i>A. amstelodami</i> , <i>A. flavus</i> , <i>A. fumigatus</i> , <i>A. nidulans</i> , <i>A. niger</i>	6 (2.6)	18 (7.3)	20 (5.9)	2 (2.1)	46 (5)

*Number (percentage) positive skin prick test responses.

†Elm tree was used at Children's Memorial Hospital in Chicago, Illinois, and olive tree was used at every other site.