

Opposite effects of *CD14*-260 on serum IgE levels in children raised in different environments

Waltraud Eder, MD,^{a,b} Walt Klimecki, PhD,^a Lizhi Yu, MD,^a Erika von Mutius, MD,^b Josef Riedler, MD,^c Charlotte Braun-Fahrlander, MD,^d Dennis Nowak, MD,^e Fernando D. Martinez, MD,^a and the Allergy and Endotoxin (ALEX) Study Team

Tucson, Ariz, Munich, Germany, Salzburg, Austria, and Basel, Switzerland

Background: Most complex diseases are the result of interactions between polymorphisms in the genome and environmental exposures.

Objective: We sought to investigate the previously reported association between a polymorphism in the promoter region of *CD14* (*CD14*-260C→T) and serum IgE levels in relation to the environment to which children are exposed.

Methods: In 624 children living in 2 rural communities in Europe, we compared total and specific serum IgE levels between the genotypes of *CD14*-260 in relation to exposure to animals and in relation to house dust endotoxin.

Results: We found that the C allele of *CD14*-260 was associated with higher levels of both total and specific serum IgE to aeroallergens in children with regular contact with pets, whereas an association in the opposite direction was found in children with regular contact with stable animals. This modifying effect of animal exposure was not explained by levels of house dust endotoxin. However, in children with high levels of house dust endotoxin, the C allele was associated with less specific IgE, independently from animal exposure.

Conclusion: Because *CD14* is a pattern recognition receptor for microbial molecules, the results suggest that the type and concentrations of such molecules present in the environment strongly determine the direction of the association between *CD14*-260 and serum markers of atopy. (J Allergy Clin Immunol 2005;116:601-7.)

Key words: Serum IgE, *CD14*-260, microbial exposure, gene-environment interaction

Most human phenotypes are the result of complex interactions between variations in the genome and environmental exposures. In many genetic studies of human disease, however, environmental factors are not concomitantly ascertained. The assumption in such studies is that most genetic variations influence the risk of disease in a unidirectional way: the affected allele increases the risk of the disease regardless of environmental exposures. On this basis, there is the expectation that discovery of genes for a disease will be replicated in at least a preponderance of corroborative studies. Recent reviews have highlighted, however, that both for association and linkage studies of complex human phenotypes, lack of widespread replication is more often the rule than the exception.¹

Although the technical flaws of many association studies might justify failure to replicate genetic findings,² other explanations are also possible. Phenotypic plasticity, which is the capacity of a genotype to produce different phenotypes depending on environmental background, has been well characterized in prokaryotes and eukaryotes.³ Recently, Remold and Lenski,⁴ for example, described opposite phenotypic effects obtained from mutations in *Escherichia coli*, depending on the growth medium in which the mutant was raised. Similar findings have been reported in plants and experimental animals³ and have been suggested for human phenotypes by Lewontin⁵ 30 years ago. However, only few studies of phenotypic plasticity are available in human subjects, most likely as a result of the difficulty in assessing well-defined environmental exposures in observational studies and to ethical limitations to experimental challenges.

The association between serum IgE levels, which are markers for the atopic status in human subjects, and a functional single nucleotide polymorphism in the promoter region of *CD14* (*CD14*-260C→T), offers an opportunity to study phenotypic plasticity.⁶ Association between the T allele for *CD14*-260 and lower levels of total or specific IgE has been reported for populations in the United States,⁷ Europe,⁸ and Asia.⁹ However, in both rural populations¹⁰ and among workers in a large mouse facility,¹¹ the T allele in *CD14*-260 was found to be associated with more atopy. What determines these apparently contradictory effects is not known, but because *CD14* encodes for a pattern recognition receptor that interacts with LPS, the endotoxin from gram-negative bacteria,¹² it is possible that the effects of *CD14*-260 on

From ^aInnate Immunity Program for Genomic Applications, Arizona Respiratory Center, University of Arizona, Tucson; ^bDr von Hauner Children's Hospital, University of Munich; ^cPaediatric Pulmonology and Allergology, Children's Hospital, Salzburg; ^dInstitute of Social and Preventive Medicine, University of Basel, Basel; and ^eInstitute and Outpatient Clinic for Occupational and Environmental Medicine, University of Munich.

Supported by the National Heart, Lung, and Blood Institute (HL66800 and HL67672); by the Bavarian Ministry for the Environment; by an Austrian grant from the Health Department of the Government of Salzburg; by the Zurich Lung Association; and by the United Bank of Switzerland.

Received for publication February 25, 2005; revised April 28, 2005; accepted for publication May 2, 2005.

Available online June 30, 2005.

Reprint requests: Fernando D. Martinez, MD, Arizona Respiratory Center, University of Arizona, 1501 N. Campbell Avenue, PO Box 245030, Tucson, AZ 85724. E-mail: fernando@arc.arizona.edu.

0091-6749/\$30.00

© 2005 American Academy of Allergy, Asthma and Immunology

doi:10.1016/j.jaci.2005.05.003

IgE might depend on microbial exposures in the environment.

To test this hypothesis, we studied the association between *CD14*–260 and serum IgE in different levels of microbial exposure in children from the Allergy and Endotoxin Study.

METHODS

Study population and exposure to microbial products

The study includes 624 children from Austria and Bavaria (Germany) who participated in the cross-sectional Allergy and Endotoxin study, as previously described.¹³ In brief, all children living on a farm in a predefined area and a random sample of children living in the same area but not raised on a farm were investigated. We used a self-administered questionnaire, which was filled out by the parents of the children, to obtain information about respiratory and allergic diseases and to enquire about exposure to factors related to rural life. As an estimate of exposure to gram-negative bacteria, endotoxin was measured in house dust. Dust samples were taken from the child's mattress,¹⁴ stored at room temperature, and shipped to a central laboratory (Munich, Germany). The content of endotoxin in all dust samples was determined by using a kinetic Limulus assay (Kinetic-QCL, BioWhittaker Inc, Walkersville, Md) and expressed in endotoxin units per square meter of mattress.¹⁵

We further used questions on exposure to various animals as a surrogate for a qualitatively broader range of exposure to microbial products because CD14 interacts with few other microbial molecules,^{16,17} apart from endotoxin. *Regular contact with stable animals* was defined as exposure at least once a week during the previous months to one or more of the following animals: cow, horse, goat, pig, or sheep. *Regular contact with pets only* was defined as having no contact with stable animals but having contact with dogs, cats, or both at least once a week during the last months. This definition includes both children exposed to pets at home and children regularly exposed to pets outside their home, for example by visiting friends, neighbors, or relatives. Children who had no regular contact with any animal during the last months were classified as having *no contact with animals*.

Total and specific serum IgE measurement

Participating children and their parents were also asked for consent to obtain a blood sample. The concentration of total serum IgE and of specific IgE against timothy grass, birch pollen, house dust mite (*Dermatophagoides pteronyssinus*), storage mite (*Lepidoglyphus destructor*), cat dander, and cow epithelium in serum was measured in a central laboratory (University Children's Hospital Berlin Charité) by using commercially available kits (CAP; Pharmacia, Uppsala, Sweden). Atopy was defined as having a specific IgE level of 3.5 IU/mL or more to any of the allergens tested, in accordance with previous reports in this population.^{13,14}

Genotyping

DNA samples were isolated from whole blood by using standard techniques. DNA concentration was determined by using fluorescence spectroscopy (PicoGreen; Molecular Probes). Genotyping of DNA samples was performed by using 5'-exonuclease (Taqman; Applied Biosystems, Foster City, Calif) assays under standard manufacturer-recommended reaction conditions (10 μ L of total volume, 0.9 μ M of PCR primers, 0.2 μ M of each probe, 20 ng of genomic DNA, and 1 $\times$ master mix; Applied Biosystems) and thermal cycling parameters (10 minutes at 95°C, 40 cycles of 15 seconds at 95°C, and 1 minute of 60°C). Thermal cycling was

performed in 9700 Viper (Applied Biosystems) instruments in 384-well plates with optical adhesive covers. Taqman reactions were analyzed in a Sequence Detection System (7900HT; Applied Biosystems) instrument. Each 384-well plate contained 15 samples for which the genotype had been determined independently by means of DNA sequencing, as well as 4 no-DNA blank wells. Fluorescence for each of the 2 probes was plotted as X,Y values, and the clusters representing genotype groups were automatically assigned by a k means-based algorithm in the SDS 2.0 software program (Applied Biosystems). Accuracy of genotypes (concordance of DNA sequencing-derived genotypes to Taqman-derived genotypes) was greater than 99.5%. The false-positive rate (blank samples being assigned to genotype clusters) was 0%.

The Taqman genotyping reaction for *CD14*–260 used the following oligonucleotides: PCR forward, AACCTAGATGC-CCTGCAGAAT; PCR reverse, ATCACCTCCCCACCTCTCTTC; Allele 1 Probe (VIC label), TACGGtCCCCCTCC; and Allele 2 Probe (FAM label), TACGGcCCCCCTC. All oligonucleotides were synthesized by Applied Biosystems. Probe synthesis included a minor groove binder moiety.

Statistical analyses

The allele frequencies of *CD14*–260 in the study population were calculated and compared with a population in Hardy-Weinberg equilibrium by using the χ^2 test with 1 *df*. Total IgE levels were log¹⁰ transformed. Endotoxin levels were divided in tertiles to explore the relationship between exposure to endotoxin, *CD14*–260, and IgE level.

Study area (Germany, Austria), age in years, and sex were considered as potential confounders and included in the statistical models. Linear regression was used to estimate adjusted mean values and SEs of total IgE for the *CD14*–260 genotypes in different categories of exposure and to test for interaction between the polymorphisms and exposures. Logistic regression was used to analyze the relationship of *CD14*–260 variants and exposure on atopic sensitization, to adjust for confounders, and to test for interaction. The likelihood ratio test was performed to test the models, including the interaction term of interest, against the model without the interaction term.

All analyses were carried out with Stata/SE 8.0 (Stata Corp, College Station, Tex), and 2-sided *P* values of less than .05 were considered significant.

The Institutional Review Board of the University of Arizona and the local Ethics Committee for Human Studies in Austria and Bavaria (Germany) approved the study, and informed written consent was obtained from the parents of the children.

RESULTS

A total of 624 children were genotyped for *CD14*–260. The allele frequency in the population was 0.51 for *CD14*–260 C, similar to the European-American reference population (proportion of the C allele 0.48; <http://innateimmunity.net>), and the genotype frequencies showed no significant deviation from a population in Hardy-Weinberg equilibrium ($\chi^2 = 0.5$, *P* > .5). Endotoxin levels from mattress samples were available for 544 children. Further characteristics of the study population and their association to endotoxin levels are given in Table I.

No association was found between *CD14*–260 and either total or specific serum IgE levels for the population

TABLE I. Characteristics of the study population in relation to exposure to endotoxin (n = 624)

	Endotoxin units/m ² mattress				P value, χ^2 test
	First tertile (98-12,494), n = 182	Second tertile (12,495-30,046), n = 181	Third tertile (30,047-530,074), n = 181	Missing, n = 80	
Study area is	65.4%	77.9%	82.3%	11.3%	<.001
Germany					
Male sex	53.3%	50.8%	50.8%	57.5%	>.5
Mean age, y (SD)	9.3 (1.48)	9.4 (1.53)	9.3 (1.50)	9.3 (1.17)	>.5*
Exposure to animals					.004
None	19.8%	13.8%	8.8%	11.3%	
Dog-cats only	23.1%	18.8%	11.6%	16.3%	
Stable animals	50.0%	59.7%	71.3%	60.0%	
Missing information	7.1%	7.7%	8.3%	12.5%	
<i>CD14/-260</i>					>.5
CC	25.3%	26.0%	25.4%	26.3%	
CT	55.0%	50.3%	48.6%	52.5%	
TT	19.8%	23.8%	26.0%	21.3%	
Total serum IgE (IU/mL)†	73.0 (58.6-90.9)	74.6 (61.0-91.2)	64.7 (52.5-79.7)	68.9 (49.3-96.2)	>.5*
Specific IgE ≥ 3.5 IU/mL‡	25.8%	20.4%	15.5%	27.5%	.051

*P value estimated by using ANOVA.

†Geometric mean (95% CI).

‡See the Methods section.

as a whole (Table II), and no association between total IgE level and the polymorphism in different levels of endotoxin exposure was seen (Fig 1). However, different levels of endotoxin significantly modified the association between specific IgE levels and *CD14/-260* variants (Fig 2). The C allele of *CD14/-260* was protective against atopy in the highest tertile of endotoxin but not in the 2 lower tertiles. Because contact to animals, particularly stable animals, is a major determinant for house dust endotoxin,^{18,19} we next explored whether the differential effects of endotoxin exposures on the association between specific IgE levels and *CD14/-260* are explained by exposure to animals. Including regular contact to various animals in the model did not explain the protective effect of the C allele against atopy in children with high exposure to endotoxin (Table III).

Moreover, regular contact with various animals itself significantly modified the association between total and specific serum IgE levels and *CD14/-260* (Figs 3 and 4). Among children who had contacts with stable animals, the C allele for *CD14/-260* was associated with less specific and less total IgE levels, whereas an association in the opposite direction was found among children with regular contact with pets only (Figs 3 and 4). In children with no contact with animals, no clear association between *CD14/-260* variants and IgE levels was detected. The interaction between *CD14/-260* and exposure to animals on total IgE and specific IgE levels remained significant after adjusting for age, sex, and endotoxin exposure (Table III).

DISCUSSION

The results of our study suggest that both levels of endotoxin in house dust and contact with various animals

TABLE II. Total serum IgE level and frequency of atopy in *CD14/-260* polymorphisms

	Total IgE (IU/mL), geometric mean (95% CI)	Atopy,* % (95% CI)
<i>CD14/-260</i>		
CC, n = 160	66.4 (54.1-81.5)	21.3 (0.15-0.28)
CT, n = 321	68.0 (58.1-79.6)	20.9 (0.16-0.25)
TT, n = 143	81.2 (62.3-105.6)	23.1 (0.16-0.30)
P value	>.2 (ANOVA, 1 df)	>.2 (χ^2 test, 2 df)

*See the Methods section.

modify the association between *CD14/-260* and serum IgE levels in children. We found the C allele of *CD14/-260* to be associated with less serum IgE in children exposed to stable animals, whereas an association in the opposite direction was found in children with regular contact with pets. A similar association between the C allele of *CD14/-260* and specific IgE levels, but not with total IgE levels, was found in children exposed to high levels of house dust endotoxin.

This is the first study that confirms in a single population apparently contradicting results reported in previous genetic studies in which environmental exposures were not measured. Baldini et al⁷ first reported the *CD14/-260* single nucleotide polymorphism and its association with total serum IgE levels in children from the Tucson Children's Respiratory Study. In that study children with positive skin test responses carrying the *CD14/-260* C allele had higher total IgE levels compared with homozygotes for the T allele. Children from this unselected birth cohort were similar with respect to ethnicity and age to those enrolled in our study population, but their urban

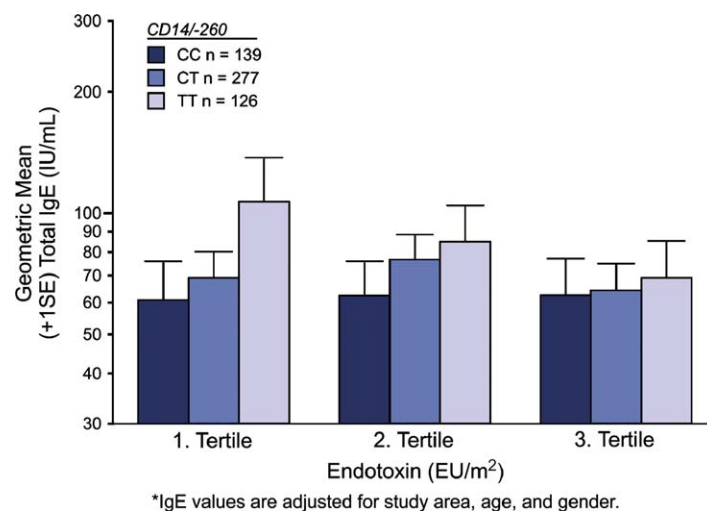


FIG 1. Total serum IgE levels in *CD14*–*260* genotypes in 3 levels of endotoxin (endotoxin units per square meter of mattress).

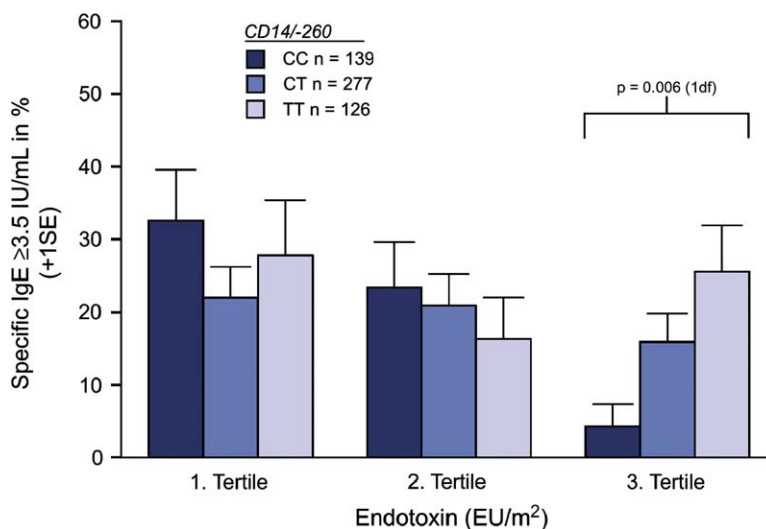


FIG 2. Specific IgE levels in *CD14*–*260* genotypes in 3 levels of endotoxin (endotoxin units per square meter of mattress).

home environment excludes regular contact with stable animals. The direction of the association between total IgE levels and *CD14*–*260* was the same as in our population when children with stable animal contacts were excluded. A very similar association was reported in adults living in The Netherlands⁸ and in children living in Hong Kong⁹ and the United Kingdom.²⁰ Conversely, no association was reported in 2 studies performed in Germany,^{21,22} and only a weak and nonsignificant association was found in a large and phenotypically well-defined Australian population.²³ Kedda et al²³ also carried out a meta-analysis including data from more than 10 populations and concluded that overall there was no association between *CD14*–*260* variants and atopy. However, the authors reported significant heterogeneity between the studies in

the association of *CD14*–*260* and atopy.²³ Although differences in study design, such as the definition used to define phenotypes, ethnicity, and the age distribution of the target population, are well-known sources of heterogeneity, the effect of exposure to microbial products might be particularly important when studying innate immunity genes. In none of the studies mentioned above was a distinction made between subjects living with or without pets or according to exposure to microbial products.

A recent report showed a significant interaction between dog ownership and *CD14*–*260* genotypes on the risk for atopic dermatitis at age 1 year.²⁴ In these infants homozygotes for the T allele were significantly less likely to have atopic dermatitis but only when a dog was kept at home, whereas no association between *CD14*–*260* and

TABLE III. Adjusted odds ratios (95% CI) for the association between *CD14*/–260 variants and specific serum IgE levels of 3.5 IU/mL or greater in relation to endotoxin levels in house dust and exposure to animals

		<i>CD14</i> /–260		
		CC	CT	TT
Endotoxin*	First tertile	1 (reference)	0.61 (0.27-1.38)	0.64 (0.22-1.88)
	Second tertile	1 (reference)	0.74 (0.29-1.86)	0.70 (0.22-2.19)
	Third tertile	1 (reference)	4.31 (0.89-20.88)‡	7.00 (1.36-36.02)§
Animal contact†	None	1 (reference)	0.52 (0.15-1.84)	0.69 (0.15-3.11)
	Dogs-cat only	1 (reference)	0.23 (0.07-0.80)§	0.25 (0.05-1.15)‡
	Stable animals	1 (reference)	1.92 (0.88-4.19)‡	2.56 (1.07-6.15)§

*Adjusted for study area, age, sex, and animal contact; *P* value for the interaction endotoxin and *CD14*/–260 variants = .070.

†Adjusted for study area, age, sex, and endotoxin; *P* value for the interaction animal contact and *CD14*/–260 variants = .018.

‡*P* = .05–.10.

§*P* < .05.

atopic dermatitis was observed among children without exposure to dogs. This finding is very similar to our observation: the negative association between the T allele of *CD14*/–260 and serum IgE levels, which, like atopic dermatitis, are also markers of the atopic status, became only apparent among children with regular contact with pets.

Contrary to what we observed in children exposed to pets, we found that the association between *CD14*/–260C and total and specific serum IgE levels was in the opposite direction among children exposed to stable animals. This finding is concordant with reports from other studies that showed a positive association between IgE responses and the T allele of *CD14*/–260 in subjects heavily exposed to animals. Among the Hutterites, for example, a population living on communal farms in South Dakota, the T allele was more often transmitted in atopic subjects than the C allele,¹⁰ and the T allele in *CD14*/–260 was associated with increased total serum IgE levels in allergic subjects working at the Jackson Laboratories in Maine.¹¹

The molecular mechanisms that explain these differential effects of environmental exposure on the association between *CD14*/–260 and markers of atopy remain to be elucidated. *CD14*/–260 has been demonstrated *in vitro* to increase transcription activity in the presence of the T allele in a monocytic cell line.²⁵ This is in concordance with the finding of a positive association between the T allele and serum-soluble CD14 found in different populations^{7,26,27} and has led to the hypothesis that carriers of the T allele, having higher CD14 expression, might be more sensitive to the putative protective effect of exposures to CD14 ligands compared with carriers of the C allele. Therefore if the development of the atopic status would vary linearly and negatively with exposure to microbial products, one would expect the T allele to be invariably protective against the development of atopy, and this is not the case. Interestingly, Eisenbarth et al²⁸ recently described a nonlinear association between exposure to endotoxin and the likelihood of development of IgE responses in an animal model of atopy. At very low levels of endotoxin exposure, mice did not have IgE

responses to a standard protocol of allergen challenge; at intermediate levels, strong responses were elicited; and at high levels of exposure, IgE responses were again suppressed. It is tempting to speculate that *CD14*/–260 shows similar and nonlinear effects on these responses depending on the genetic variation and the concentrations of endotoxin in the environment.

There is another possible and not mutually exclusive explanation for our findings. Recent reports indicate that CD14 is part of a pattern recognition receptor complex that not only interacts with endotoxin but also with other microbial components, such as lipopeptides, lipoteichoic acid, and peptidoglycan, and with a variety of lipid-containing molecules, such as ceramid, gangliosides, and glycosphingolipids.²⁹ It is possible that molecules other than endotoxin but also associated with exposure to animals contribute to the observed association between *CD14*/–260 and IgE levels in our population. Children with contact to pets could be exposed to microbial products quite different from those to which children with contact to stable animals are exposed. The lack of any modification of the association between total serum IgE levels and *CD14*/–260 by levels of endotoxin, in contrast to the strong effect modification by exposure to various animals, supports this point of view. Furthermore, the interaction between animal contact and *CD14*/–260 as determinants of either total IgE or specific IgE levels was not explained by levels of endotoxin. One other putative CD14 ligand apart from endotoxin is peptidoglycan, and a major component of this molecule, muramic acid, has been measured in a subsample of the Allergy and Endotoxin Study population.³⁰ However, levels of muramic acid are available for only 377 of 624 children genotyped for *CD14*/–260 and are highly correlated with levels of endotoxin (*r* = 0.7), as previously reported.³⁰ The sample size of this study population therefore does not allow disentangling the observed associations between IgE levels and *CD14*/–260 by levels of endotoxin to those by levels of muramic acid, and further studies will be needed to address this issue.

Contrary to total IgE, both high levels of house dust endotoxin and regular contact with stable animals modified

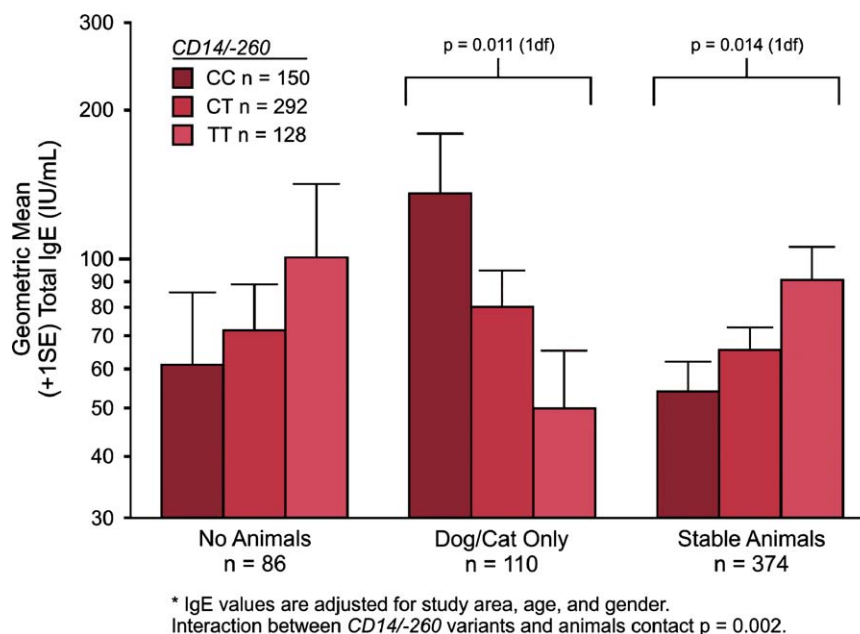


FIG 3. Total serum IgE levels in *CD14/-260* genotypes in relation to exposure to animals.

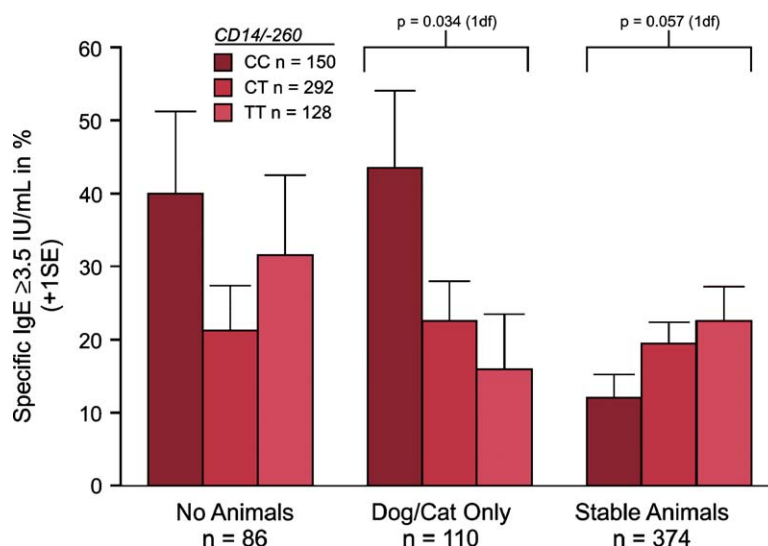


FIG 4. Specific IgE levels in *CD14/-260* genotypes in relation to exposure to animals.

the direction of the association between *CD14/-260* and specific IgE levels. It thus appears that in carriers of the C allele, stable animal contact prevents IgE production regardless of its nature, whereas exposure to high endotoxin levels suppresses the production of specific IgE to aeroallergens only. The underlying mechanisms by which endotoxin and exposure to animals influence IgE production in relation to *CD14/-260* are therefore likely to be different.

Our findings might have important implications for genetic studies of complex diseases because the genetic

component only became apparent when the environmental exposures were considered. It is plausible to surmise that a genetic component of atopic phenotypes in relation to *CD14/-260* should be found in some studies but not in others depending on the level of environmental exposure present in the population under study. In this situation only studies with similar exposures will appear to replicate each other's results. If the pattern of marked phenotypic plasticity observed for IgE responses is also present for other phenotypes, such plasticity might be an important determinant of genetic heterogeneity in human subjects

and thus a major cause underlying lack of replication between genetic studies.

We thank the collaborators of the Allergy and Endotoxin Study Team: Soyoun Maisch, Rudi Schierl, David Carr (Munich Germany), Marco Waser (Basel, Switzerland), Roger Lauener, Felix Sennhauser (Zürich, Switzerland), Mynda Schreuer, Gerd Oberfeld (Salzburg, Austria), Harald Renz, Udo Herz (Marburg Germany), Albrecht Bufer (Bochum, Germany), Otto Holst (Borstel, Germany), and Donata Vercelli, MD, for her helpful suggestions.

REFERENCES

- Hirschhorn JN, Lohmueller K, Byrne E, Hirschhorn K. A comprehensive review of genetic association studies. *Genet Med* 2002;4:45-61.
- In search of genetic precision. *Lancet* 2003;361:357.
- Schlichting CD, Pigliucci M. Reaction norms and phenotypic plasticity. In: *Phenotypic evolution: a reaction norm perspective*. Sunderland (MA): Sinauer Associates; 1998. p. 51-84.
- Remold SK, Lenski RE. Pervasive joint influence of epistasis and plasticity on mutational effects in *Escherichia coli*. *Nat Genet* 2004;36:423-6.
- Lewontin RC. The analysis of variance and the analysis of causes. *Am J Hum Genet* 1994;26:400-11.
- Vercelli D. Learning from discrepancies: CD14 polymorphisms, atopy and the endotoxin switch. *Clin Exp Allergy* 2003;33:153-5.
- Baldini M, Lohman C, Halonen M, Erickson RP, Holt PG, Martinez FD. A polymorphism in the 5' flanking region of the CD14 gene is associated with circulating soluble CD14 levels and with total serum immunoglobulin E. *Am J Respir Cell Mol Biol* 1999;20:976-83.
- Koppelman GH, Reijmerink NE, Stine OC, Howard TD, Whittaker PA, Meyers DA, et al. Association of a promoter polymorphism of the CD14 gene and atopy. *Am J Respir Crit Care Med* 2001;163:965-9.
- Leung TF, Tang NLS, Sung YM, Li AM, Kong GWK, Chan HIS, et al. The C-159T polymorphism in the CD14 promoter is associated with serum total IgE concentration in atopic Chinese children. *Pediatr Allergy Immunol* 2003;14:255-60.
- Ober C, Tsalenko A, Parry R, Cox NJ. A second-generation genomewide screen for asthma-susceptibility alleles in a founder population. *Am J Hum Genet* 2000;67:1154-62.
- Amelung PJ, Weisch DG, Xu J, Paigen B, Meyers DA, Bleecker ER. A polymorphism in CD14 is associated with high IgE levels in a population with laboratory animal allergy [abstract]. *Am J Respir Crit Care Med* 2000;161:A927.
- Heumann D, Roger T. Initial responses to endotoxin and gram-negative bacteria. *Clin Chim Acta* 2002;323:59-72.
- Riedler J, Braun-Fahrlander C, Eder W, Schreuer M, Waser M, Maisch S, et al. Exposure to farming in early life and development of asthma and allergy: a cross-sectional survey. *Lancet* 2001;358:1129-33.
- Braun-Fahrlander C, Riedler J, Herz U, Eder W, Waser M, Grize L, et al. Environmental exposure to endotoxin and its relation to asthma in school-age children. *N Engl J Med* 2002;347:869-77.
- Hollander A, Heederik D, Versloot P, Douwes J. Inhibition and enhancement in the analysis of airborne endotoxin levels in various occupational environments. *Am Ind Hyg Assoc J* 1993;54:647-53.
- Dziarski R. Recognition of bacterial peptidoglycan by the innate immune system. *Cell Mol Life Sci* 2003;60:1793-804.
- Lotz S, Aga E, Wilde I, van Zandbergen G, Hartung T, Solbach W, et al. Highly purified lipoteichoic acid activates neutrophil granulocytes and delays their spontaneous apoptosis via CD14 and TLR2. *J Leukoc Biol* 2004;75:467-77.
- Waser M, Schierl R, von Mutius E, Maisch S, Carr D, Riedler J, et al. Determinants of endotoxin levels in living environments of farmers' children and their peers from rural areas. *Clin Exp Allergy* 2004;34:389-97.
- Gehring U, Bischof W, Borte M, Herbarth O, Wichmann HE, Heinrich J, et al. Levels and predictors of endotoxin in mattress dust samples from East and West German homes. *Indoor Air* 2004;14:284-92.
- Gao PS, Mao XQ, Baldini M, Roberts MH, Adra CN, Shirakawa T, et al. Serum total IgE levels and *CD14* on chromosome 5q31. *Clin Genet* 1999;56:164-5.
- Sengler C, Haider A, Sommerfeld C, Lau S, Baldini M, Martinez F, et al. Evaluation of the CD14 C-159 T polymorphism in the German Multicenter Allergy Study cohort. *Clin Exp Allergy* 2003;33:166-9.
- Kabesch M, Hasemann K, Schickinger V, Tzotcheva I, Bohnert A, Carr D, et al. A promoter polymorphism in the CD14 gene is associated with elevated levels of soluble CD14 but not with IgE or atopic diseases. *Allergy* 2004;59:520-5.
- Kedda MA, Lose F, Duffy D, Bell E, Thompson PJ, Upham J. The CD14 C-159T polymorphism is not associated with asthma or asthma severity in an Australian adult population. *Thorax* 2005;60:211-4.
- Gern JE, Reardon CL, Hoffman S, Nicolae D, Li Z, Roberg KA, et al. Effects of dog ownership and genotype on immune development and atopy in infancy. *J Allergy Clin Immunol* 2004;113:307-14.
- LeVan TD, Bloom JW, Bailey TJ, Karp CL, Halonen M, Martinez FD, et al. A common single nucleotide polymorphism in the CD14 promoter decreases the affinity of Sp protein binding and enhances transcriptional activity. *J Immunol* 2002;167:5838-44.
- Karhukorpi J, Yan Y, Niemela S, Valtonen J, Koistinen P, Joensuu T, et al. Effect of CD14 promoter polymorphism and *H. pylori* infection and its clinical outcomes on circulating CD14. *Clin Exp Immunol* 2002;128:326-32.
- Obana N, Takahashi S, Kinouchi Y, Negoro K, Takagi S, Hiwatashi N, et al. Ulcerative colitis is associated with a promoter polymorphism of lipopolysaccharide receptor gene, CD14. *Scand J Gastroenterol* 2002;37:699-704.
- Eisenbarth SC, Piggott DA, Huleatt JW, Visintin I, Herrick CA, Bottomly K. Lipopolysaccharide-enhanced, toll-like receptor 4-dependent T helper cell type 2 responses to inhaled antigen. *J Exp Med* 2002;196:1645-51.
- Schmitz G, Orso E. CD14 signalling in lipid rafts: new ligands and co-receptors. *Curr Opin Lipidol* 2002;13:513-21.
- Van Strien RT, Engel R, Holst O, Bufer A, Eder W, Waser M, et al. Microbial exposure of rural school children, as assessed by levels of N-acetyl-muramic acid in mattress dust, and its association with respiratory health. *J Allergy Clin Immunol* 2004;113:860-7.