

Mouse allergen exposure and immunologic responses: IgE-mediated mouse sensitization and mouse specific IgG and IgG4 levels

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Background: Although there is evidence that contact with mice is associated with IgE-mediated mouse sensitization and mouse specific antibody responses, the exposure-response relationships remain unclear.

Objective: To determine whether IgE-mediated mouse sensitization and mouse specific IgG (mIgG) and mIgG4 levels increase with increasing Mus m 1 exposure.

Methods: One hundred fifty-one workers at a mouse research and production facility were studied. Exposure assignments were made by linking participants to airborne Mus m 1 concentrations in their respective work areas. Cumulative exposure was estimated by multiplying airborne Mus m 1 concentration by duration of employment. Serum mIgG and mIgG4 levels were quantified by antigen-binding assays, and IgE-mediated mouse sensitization was evaluated by skin prick testing (SPT).

Results: Prevalence rates of mouse SPT sensitivity and of high levels of mIgG and mIgG4 were increasingly higher by quintiles of increasing cumulative exposure ($P < .01$ for SPT, mIgG, and mIgG4). After adjusting for age, sex, and atopy, the log odds ratio (OR) of having positive mouse SPT results was linearly related to cumulative exposure ($r^2 = 0.87$), as was the log OR of having a high mIgG level ($r^2 = 0.86$). Quintile of cumulative exposure was an independent predictor of both SPT sensitivity (OR, 1.7; 95% confidence interval, 1.2–2.5) and a high mIgG level (OR, 1.7; 95% confidence interval, 1.2–2.4).

Conclusions: IgE-mediated mouse sensitization and mIgG and mIgG4 levels were related to cumulative exposure in a dose-dependent manner. Thus, strategies to prevent allergy to mice should remain focused on reducing mouse allergen exposure.

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INTRODUCTION

Laboratory rodent allergy significantly affects the health and productivity of laboratory rodent workers.^{1–3} Although contact with mice has been established as a risk factor for the development of IgE-mediated mouse sensitization,⁴ it remains unclear whether sensitization risk increases as exposure increases. Mouse allergen exposure has also been shown to be almost ubiquitous in inner-city homes of asthmatic children,⁵ and exposure to high levels of Mus m 1 in household settled dust is associated with mouse skin prick test (SPT) sensitivity. Although a few studies^{6–8} have provided evidence to

support a dose-response relationship between rat allergen exposure and IgE-mediated rat sensitization, most studies^{2,9} have only demonstrated that exposure in general is related to sensitization for rodent allergens. For mouse allergen in particular, no dose-response relationship has been found. Evidence of such a relationship would be critical in establishing goals for reducing mouse allergen exposure, with the ultimate aim of reducing the risk of IgE-mediated mouse sensitization.

Nonallergic antibody responses, including IgG and IgG4, have been associated with the development of tolerance.^{10,11} Allergen specific IgG and IgG4 levels increase with immunotherapy¹² and, in the setting of natural exposure to Fel d 1, have also been shown to be inversely related to IgE-mediated sensitization to cats.¹³ Studies of occupational mouse exposure and allergen specific IgG suggest that exposure to mice increases the risk of developing mouse specific IgG (mIgG),⁴ but whether elevated levels of mIgG are associated with tolerance to mouse allergen remains unknown.

To determine whether mouse-specific immune responses are related to Mus m 1 exposure in a dose-dependent manner, we conducted a cross-sectional study of employees of a mouse research and production facility and examined mIgG and mIgG4 levels and IgE-mediated mouse sensitization.

METHODS

Study Design and Study Population

Employees of a mouse research and production facility were recruited through advertisements and underwent SPT, had a

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blood sample taken, and completed a questionnaire during a single study visit. All employees of The Jackson Laboratory were eligible to participate in the study, and participants provided written informed consent. The protocol was approved by the institutional review boards at the Johns Hopkins School of Medicine and The Jackson Laboratory. Of 1,200 employees, 284 (24%) participated. Of these, 151 had valid SPT results, serum samples available for mouse-specific antibody measurements, and exposure measurements. This subgroup was similar to the other participants in terms of age, duration of employment, smoking status, atopic disease history, and IgE-mediated mouse sensitization.

mIgG and mIgG4

Levels of mIgG and mIgG4 were measured in serum samples using a solid-phase antigen-binding assay as previously described.¹⁴ Briefly, for the detection of IgG, serum samples were incubated overnight with protein G to determine IgG1, IgG2, IgG3, and IgG4 levels (CNBr-activated Sepharose 4B; Pharmacia Diagnostics, Uppsala, Sweden). Per test, 1 to 20 μ L of serum was added to 500 μ g of Sepharose in a total volume of 800 μ L (phosphate-buffered saline solution/0.3% human serum albumin/0.1% Tween 20) together with radiolabeled mouse urinary protein 8 (a Mus m 1 isoform) for detection.¹⁵ After washing, the amount of bound radioactivity was measured and read from a standard curve, a human pooled reference serum. Mouse specific IgG4 levels were determined using anti-IgG4 solid phase (CNBr-activated Sepharose 4B). Again, 1 to 20 μ L of serum was added per test to 500 μ g of Sepharose in a total volume of 800 μ L (phosphate-buffered saline solution/0.3% human serum albumin/0.1% Tween 20). After overnight incubation, the samples were washed and incubated overnight with radiolabeled mouse urinary protein 8. The samples were washed, and the amount of bound radioactivity was measured. The previously mentioned reference serum was used as a standard. The results are expressed in arbitrary units per milliliter of serum (AU/mL), and the detection limit of the antigen-binding assay was 0.5 AU/mL for IgG and 2.0 AU/mL for IgG4.

SPT and CAP Radioallergosorbent Testing

Skin tests to cat, dog, *Dermatophagoides pteronyssinus*, *Dermatophagoides farinae*, ragweed, grass, oak, *Alternaria*, *Aspergillus*, and rat, mouse, hamster, guinea pig, and rabbit epithelia were performed using full-strength glycerinated extracts and a disposable allergy SPT device (MultiTest; Lincoln Diagnostics, Decatur, IL). Skin test results were considered positive if the orthogonal wheal diameter was at least 3 mm greater than the negative control and at least half the size of the histamine control. Serum levels of mouse allergen specific IgE were quantified using the ImmunoCap 100 System and mouse urine immunocaps (Pharmacia Diagnostics). A mouse urine CAP radioallergosorbent test (RAST) value of 0.35 kU_A/L or greater was considered positive.

Questionnaire

Participants completed a self-administered questionnaire composed of the validated American Thoracic Society respiratory

symptom questionnaire¹⁶ and a questionnaire adapted from the Collaborative Study on the Genetics of Asthma questionnaire.¹⁷ A participant met the criteria for asthma if he or she had ever had asthma confirmed by a physician. A participant was considered to have self-reported allergic rhinitis if he or she responded positively to the question, "Have you ever had hay fever?"

Exposure Evaluation

Air samples were collected throughout The Jackson Laboratory between January 2000 and April 2002. Each area was sampled Monday through Thursday using a 20-L/min air-sampling pump and an impactor (Air Diagnostics, Harrison, ME). A stainless steel plate captured particles larger than 10 μ m in diameter, and a 2- μ m Teflon filter (Chester LabNet, Tigard, OR) captured particles smaller than 10 μ m. Protein was extracted from the filters using a standardized protocol, and Mus m 1 was quantified by sandwich enzyme-linked immunosorbent assay using immunosorbent sheep-purified anti-Mus m 1.¹⁸ Purified Mus m 1 was used for the calibration curve.¹⁹ Areas that were sampled, in addition to mouse rooms, included the safety building, the computing building, customer service, the laboratory animal health building, the warehouse, clean supply areas, offices, research laboratories, and the cafeteria. The safety building, computing building, and warehouse are separate structures from buildings containing mice and had undetectable mouse allergen. Customer service and the clean supply areas, although housed in the same buildings as mice, had undetectable mouse allergen levels. The 4-day average airborne Mus m 1 level was taken as the average room exposure. Study participants identified a primary area of work, and the Mus m 1 level in that room was used as a measure of current exposure. To take duration of exposure into account, cumulative exposure was estimated as average Mus m 1 concentration multiplied by the duration of employment, with resulting units of nanograms per cubic meter-years (ng/m³-y). The assumption that participants' current exposure reflected average exposure throughout the duration of their employment was supported by the fact that 86% of current study participants had held no more than 2 different jobs. In addition, for 90% of those who had held 2 different jobs, both jobs entailed handling mice.

Statistical Analysis

All analyses were performed using STATA 7.0 (STATA Corp, College Station, TX). Categorical variables were compared using χ^2 analysis, and continuous variables were compared using the Student *t* test or the Mann-Whitney *U* test, depending on the distribution of the data. Atopy was defined as at least 1 positive SPT result aside from mouse. Both current and cumulative Mus m 1 exposure were divided into quintiles. Exposure-response relationships between Mus m 1 exposure and prevalence rates of IgE-mediated mouse sensitization and high mIgG and mIgG4 levels were analyzed using the Cuzick nonparametric test for trend.²⁰ Levels of mIgG and mIgG4 greater than the 75th percentile for the study population were considered to be high antibody

responses. This cutoff value was chosen arbitrarily because no threshold level has been established and results from these assays are not comparable from laboratory to laboratory.

Multivariate logistic regression was used to test the hypothesis that the log odds ratio (OR) of having a positive mouse skin prick test (SPT) was linearly related to cumulative mouse allergen exposure while controlling for the possible confounders of age, sex, and atopy. The adjusted OR (on a log scale) of a positive mouse SPT result was plotted against the midpoint of each quintile of cumulative exposure. The final multivariate logistic regression model resulted in a coefficient for quintile of cumulative mouse allergen exposure that represents the change in the log odds of a positive mouse SPT result with each increase in quintile of cumulative exposure. The same analytic approach was used when a positive mouse urine CAP RAST result and mIgG were the outcome variables of interest.

To test the hypothesis that the relationship between the log OR of a positive mouse SPT result and cumulative mouse allergen exposure is an inverted "U" (a pattern consistent with high-dose tolerance), a quadratic cumulative exposure term was added to the final multivariate model, and a likelihood ratio test was performed to determine whether this quadratic model was a better fit than the linear model. The threshold selected for statistical significance was $P < .05$ for all analyses.

RESULTS

Study Population

Animal caretakers, research technicians, and administrative and support personnel made up 90.0% of the 151 subjects. The average age of the study population was 37.9 years. Almost half had college degrees, 66.9% were female, and 97.4% were white (Table 1). Seventy percent of the participants currently handled mice. The median duration of employment was 3 years. Forty-six percent of the participants were atopic, and 23.2% were current smokers. Physician-diagnosed asthma was reported by 17.9%, and the prevalence rate of self-reported allergic rhinitis was 20.7%. Almost 9% of the participants had received immunotherapy, and 2% had been hospitalized for asthma. Almost 18% (27/151) of the participants worked in areas where mouse allergen was below detection, and 18.5% (28/151) had never handled mice. Of the 33 participants with mouse SPT sensitivity, 18 (54.5%) reported nasal or ocular symptoms and 10 (30.3%) reported lower respiratory tract symptoms with exposure to mice. In contrast, of the 118 participants without mouse SPT sensitivity, 11 (9.3%) reported nasal or ocular symptoms and 6 (5.1%) reported lower respiratory tract symptoms. The differences in reported symptoms between the SPT positive and negative groups were statistically significant for both nasal or

Table 1. Study Population Characteristics and Risk Factors for Mouse Sensitization

Characteristic	Study population (n = 151)*	Positive mouse SPT results*	Crude OR (95% CI)
Sex			
Female	104 (66.9)	28 (27.7)	3.5 (1.2–9.6)
Male	47 (33.1)	5 (10.0)	1.0
Education			
College degree	73 (48.3)	19 (26.0)	1.6 (0.7–3.5)
No college degree	78 (51.7)	14 (18.0)	1.0
Smoking status			
Current smoker	35 (23.2)	8 (22.9)	1.1 (0.4–2.7)
Nonsmoker	116 (76.8)	25 (21.6)	1.0
Race			
White	147 (97.4)	32 (21.8)	0.8 (0.1–8.3)
Nonwhite	4 (2.6)	1 (25.0)	1.0
Mouse contact			
Mouse handler	106 (70.2)	26 (24.5)	1.8 (0.7–4.4)
Non-mouse handler	45 (29.8)	7 (15.6)	1.0
Job category			
Administrative/support	33 (21.9)	4 (12.1)	0.4 (0.1–1.3)
Scientist	6 (4.0)	1 (16.7)	0.7 (0.1–6.3)
Animal caretaker	50 (33.1)	9 (18.0)	0.7 (0.3–1.7)
Laboratory technician	53 (35.0)	19 (35.9)	3.4 (1.5–7.4)
Other	9 (6.0)	0 (0)	—
Allergic characteristics			
Atopic	70 (46.4)	28 (40.0)	10.1 (3.6–28.2)
Asthma	27 (17.9)	12 (44.4)	3.9 (1.6–9.6)
Self-reported allergic rhinitis	31 (20.7)	10 (32.3)	2.0 (0.8–4.8)
Immunotherapy, ever	13 (8.7)	4 (30.8)	1.7 (0.5–5.8)
Asthma hospitalization, ever	3 (2.0)	2 (66.7)	7.5 (0.7–86.0)

Abbreviations: CI, confidence interval; OR, odds ratio; SPT, skin prick test.

* Data are given as number (percentage).

Table 2. Study Population Characteristics by Mouse Sensitization Status

Characteristic	Total population (n = 151)	Mouse SPT result		P value
		Negative (n = 118)	Positive (n = 33)	
Age (y), mean $\pm$ SD	37.9 $\pm$ 10.9	37.9 $\pm$ 10.8	37.9 $\pm$ 11.5	.99
Years worked, median (IQR)*	3 (1–8)	2 (1–7)	5 (2–15)	.02
Mouse IgG (AU/mL), median (IQR)	5 (2–12)	4 (2–7)	28 (6–227)	<.001
Mouse IgG4 (AU/mL), median (IQR)	BD (BD–3)	BD (BD–2)	10 (BD–27)	<.001
Current Mus m 1 (ng/m ³), median (IQR)	0.13 (0.04–0.30)	0.13 (0.04–0.30)	0.14 (0.13–0.42)	.12
Cumulative Mus m 1 (ng/m ³ -y), median (IQR)	0.29 (0.04–1.3)	0.27 (0.03–0.90)	1.16 (0.28–3.4)	<.001

Abbreviations: AU, arbitrary unit; BD, below detection; IQR, interquartile range; SPT, skin prick test.

* Duration of employment at The Jackson Laboratory.

ocular symptoms and lower respiratory tract symptoms ($P < .001$ for both). The median current exposure concentration of Mus m 1 was 0.13 ng/m³, and the median cumulative exposure was 0.29 ng/m³-y. The median mIgG level was 5 AU/mL, and the median mIgG4 level was below detection, with approximately one fourth of the participants having detectable levels of mIgG4 (Table 2).

Risk Factors for Mouse Sensitization

Of the 151 study participants, 33 (21.9%) had positive SPT results to mouse, and 16 (10.8%) of 148 had positive mouse CAP RAST results. Current Mus m 1 exposure was not associated with mouse SPT sensitivity, but cumulative exposure was (median, 1.16 and 0.27 ng/m³-y for the sensitized and unsensitized groups, respectively; $P < .001$). Longer duration of employment was also associated with SPT sensitivity (median, 5 and 2 years for the sensitized and unsensitized groups, respectively; $P = .02$; Table 2).

There were no differences in the prevalence of mouse SPT sensitivity by race, education, or smoking status (Table 1). Females had a more than 3-fold greater risk of mouse sensitization. Laboratory technicians were 3.5 times more likely to have a positive mouse SPT result than workers in other job categories, but no significant risk was associated with any other job category. Atopy was associated with a more than 10-fold increased risk of mouse SPT sensitivity. A history of asthma was also associated with a positive mouse SPT result (OR, 3.9; 95% confidence interval [CI], 1.6–9.6), but self-

reported allergic rhinitis was not (OR, 2.0; 95% CI, 0.8–4.8). Hospitalization for asthma was associated with a more than 7-fold risk of a positive mouse SPT result, but this finding was not statistically significant (OR, 7.5; 95% CI, 0.7–86.0).

Allergic sensitization to specific allergens was, in general, associated with a statistically significant increased risk of mouse SPT sensitivity, although the associations were no longer significant after controlling for atopy, except for rodent/lagomorph sensitivity (Table 3). Sensitization to rat, rabbit, guinea pig, or hamster was associated with a more than 4-fold increase in risk of mouse SPT sensitivity after controlling for atopy (OR, 4.2; 95% CI, 1.5–12.0).

Mouse specific IgG levels were higher in the positive SPT result group. The median mIgG level was 4 AU/mL among those with a negative mouse SPT result and 28 AU/mL among those with a positive mouse SPT result ($P < .001$) (Table 2); the median mIgG4 level was below detection and 10 AU/mL, respectively ($P < .001$). A history of domestic mouse exposure was not associated with IgE-mediated mouse sensitization (data not shown).

Mouse Sensitization, Mouse-Specific Antibodies, and Mus m 1 Exposure

Cumulative, but not current, Mus m 1 exposure was associated with IgE-mediated mouse sensitization and mIgG and mIgG4 levels in a dose-dependent manner (Fig 1). The prevalence of mouse SPT sensitivity increased from a low of 9.4% in the lowest quintile to a high of 44.4% in the highest quintile of

Table 3. Association of Specific Skin Prick Test Sensitivities with Mouse Skin Prick Test Sensitivity

Allergen(s)	OR (95% CI)	
	Crude	Adjusted*
Cat or dog	5.6 (2.2–14.3)	2.1 (0.8–5.8)
Rodents/lagomorphs (rat, rabbit, guinea pig, or hamster)	10.1 (3.8–26.5)	4.2 (1.5–12.0)†
Dust mite (<i>Dermatophagoides farinae</i> or <i>Dermatophagoides pteronyssinus</i>)	4.4 (1.9–9.8)	1.1 (0.4–2.9)
Pollens (grass, oak, or ragweed)	3.8 (1.7–8.8)	1.2 (0.4–3.0)
Molds (<i>Alternaria</i> or <i>Aspergillus</i>)	4.3 (1.4–13.2)	1.7 (0.5–5.4)

Abbreviations: CI, confidence interval; OR, odds ratio.

* Adjusted for atopy.

† Statistically significant.

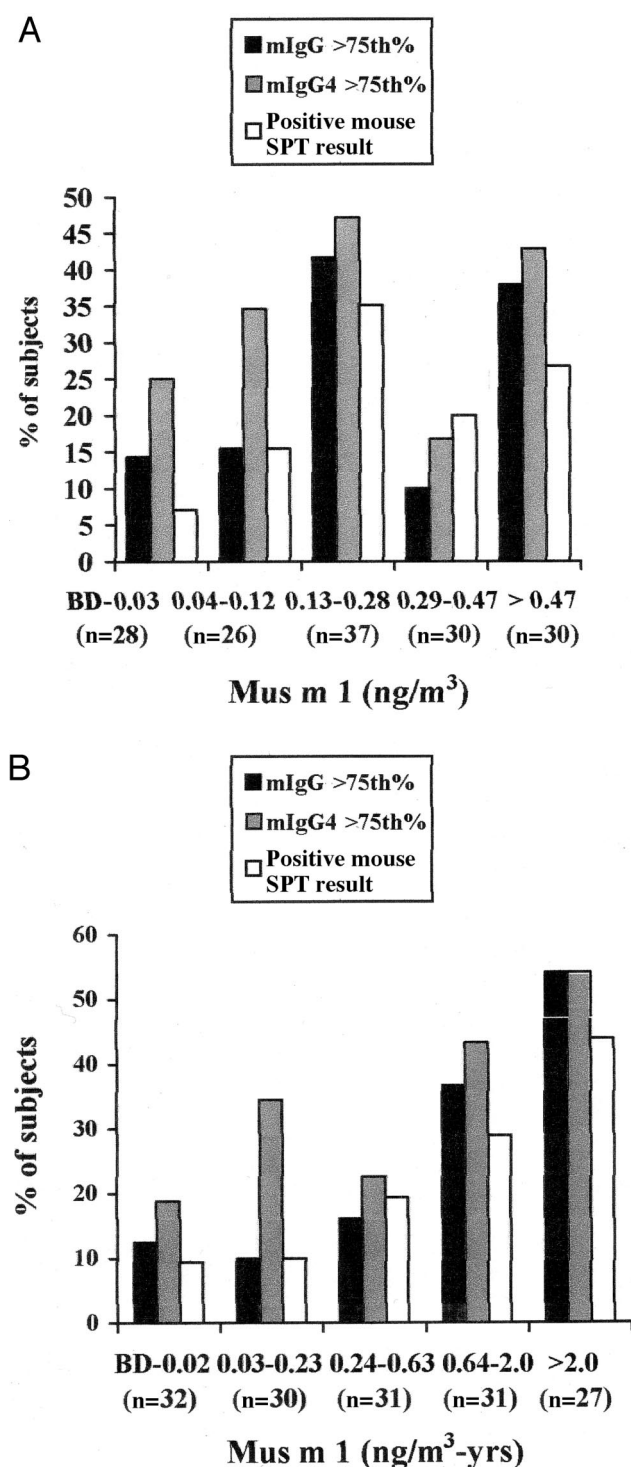


Figure 1. Relationship between Mus m 1 exposure and elevated mouse specific IgG (mIgG) and mIgG4 levels and mouse sensitization. A, Current Mus m 1 exposure (sensitization, $P = .08$; mIgG, $P = .56$; and mIgG4, $P = .11$). B, Cumulative Mus m 1 exposure (sensitization, mIgG, and mIgG4, $P < .01$ for all). Statistical analysis was performed using the Cuzick test for trend. BD indicates below detection; SPT, skin prick test.

cumulative exposure ($P < .01$). High mIgG levels increased with higher quintiles of cumulative exposure (12.5%, 10.0%, 16.1%, 36.7%, and 53.9%, respectively; $P < .01$). Likewise, high mIgG4 levels increased with higher quintiles of cumulative exposure (18.8%, 34.5%, 22.6%, 43.3%, and 53.9%, respectively; $P < .01$). Similar increasing monotonic relationships were seen when the cutoff values for high mIgG and mIgG4 levels were set at the median (data not shown). Cumulative Mus m 1 exposure was also associated with having a positive mouse CAP RAST result in a dose-dependent manner (prevalence, 3.1%, 6.7%, 6.5%, 16.1%, and 25.0% in each quintile, respectively; $P = .01$). In addition, CAP RAST values increased with increasing quintiles of cumulative exposure, and the trend was statistically significant (data not shown).

Changes in Job Category

We examined whether changes in job category were related to mouse SPT sensitivity status. Of the 33 participants who had a positive mouse SPT result, 23 (69%) had held either only 1 job or, if they had held a previous job, both jobs were in the same job category. Similarly, of the 118 participants who did not have a positive mouse SPT result, 76% had held either only 1 job or, if they had held a previous job, both jobs were in the same job category ($P = .44$). We also examined the rates of workers moving from research, animal care, or scientist positions to an administrative or support job and found that the rates were similar in the SPT positive group vs the SPT negative group (2 of 33 vs 4 of 118, respectively; $P = .49$).

Multivariate Logistic Regression

After controlling for age, sex, and atopy, there was a strong linear relationship between the log OR of having a positive mouse SPT result and cumulative Mus m 1 exposure (Fig 2A). In the final multivariate logistic regression model, this relationship corresponded to an OR of 1.7 (95% CI, 1.2–2.5) of a positive mouse SPT result for each increase in quintile of cumulative mouse allergen exposure (Table 4). Female sex (OR, 3.7; 95% CI, 1.2–12.0) and atopy (OR, 11.3; 95% CI, 3.8–33.6) were also independent predictors of mouse sensitization. Similarly, there was a 70% increase in the risk of having a positive mouse CAP RAST result with each increase in quintile of cumulative exposure (OR, 1.7; 95% CI, 1.1–2.7) after adjusting for age, sex, and atopy. A linear relationship was also found between the adjusted log OR of having a high mIgG level and cumulative Mus m 1 exposure (Fig 2B), which corresponded to an OR of 1.7 (95% CI, 1.2–2.4) of having a high mIgG level with each increase in quintile of cumulative exposure. Atopy (OR, 2.4; 95% CI, 1.0–5.5) was also an independent predictor of a high mIgG level. A model including a quadratic cumulative exposure term did not provide a better fit of the data by likelihood ratio testing ($P = .24$).

DISCUSSION

This study provides evidence for an increasingly monotonic relationship between cumulative Mus m 1 exposure and IgE-mediated mouse sensitization and mIgG and mIgG4 levels. Our findings suggest that long-term exposure to mice

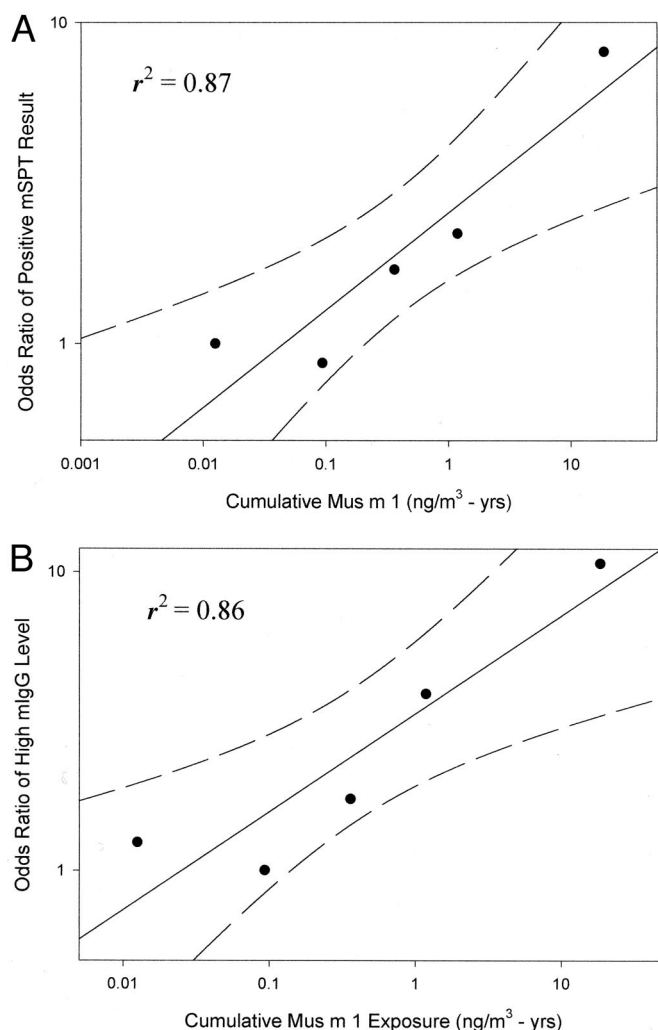


Figure 2. Adjusted odds ratios (log scale) of a positive mouse skin prick test (mSPT) result (A) and high mouse specific IgG levels (B) plotted against the midpoint of each quintile of cumulative Mus m 1 exposure. The solid line is the linear regression line, and the dashed lines represent the 95% confidence interval for the line.

is a significant risk factor for IgE-mediated sensitization and, furthermore, may drive mouse specific non-IgE antibody responses.

Although previous studies^{2,4,9} have demonstrated an increased risk of sensitization among individuals with greater rodent contact, these studies used approximations of exposure, such as job descriptions or hours per week of rodent contact. We included airborne Mus m 1 concentration and a measure of exposure duration in our exposure metric, resulting in an estimate of cumulative exposure. This exposure metric is consistent with the observation that duration and intensity of exposure play important roles in sensitization, and it may be more biologically relevant than cross-sectional exposure measurements. In fact, a previous study²¹ found that

Table 4. Mus m 1 Exposure and Risk of IgE-Mediated Mouse Sensitization and mIgG

	OR (95% CI)	
	Unadjusted	Adjusted*
Mouse SPT		
Age (years)	1.0 (0.9–1.0)	1.0 (0.9–1.0)
Atopy	12.7 (4.9–33.3)	11.3 (3.8–33.6)
Sex (female vs male)	3.5 (1.2–9.6)	3.7 (1.2–12.0)
Cumulative exposure†	1.8 (1.3–2.4)	1.7 (1.2–2.5)
Mouse urine CAP RAST		
Age	1.0 (1.0–1.1)	1.0 (0.9–1.1)
Atopy	10.1 (2.2–46.3)	9.2 (2.0–43.4)
Sex	1.1 (0.4–3.5)	1.0 (0.3–3.5)
Cumulative exposure†	1.8 (1.2–2.8)	1.7 (1.1–2.7)
mIgG		
Age	1.0 (1.0–1.1)	1.0 (1.0–1.1)
Atopy	2.7 (1.2–5.7)	2.4 (1.0–5.5)
Sex	1.5 (0.7–3.4)	1.3 (0.5–3.2)
Cumulative exposure†	1.9 (1.4–2.6)	1.7 (1.2–2.4)

Abbreviations: CI, confidence interval; mIgG, mouse specific IgG; OR, odds ratio; RAST, radioallergosorbent test; SPT, skin prick test.

* Adjusted for age, atopy, sex, and cumulative exposure.

† Quintiles.

time until the development of laboratory animal allergy symptoms was shorter for higher-intensity exposure. Two other published studies^{5,6} have used a time-multiplied exposure measure as an exposure metric for laboratory rat workers. In these studies, however, airborne levels of Rat n 1 were multiplied by the average number of hours per week of rat exposure as a measure of average weekly exposure rather than by the full duration of exposure. We multiplied room air Mus m 1 concentration by duration of employment as a surrogate for cumulative exposure and found this measure of exposure to be a stronger predictor of sensitization risk than either current exposure or duration of exposure alone. Our methods assume, much like the pack-year concept of cigarette smoking, that individuals exposed to 1 ng/m³ of Mus m 1 for more than 2 years are at the same risk as those exposed to 2 ng/m³ for 1 year. If this assumption is correct, strategies to decrease average room air levels of Mus m 1 by 50% would double the time it takes for workers to develop IgE-mediated sensitization to mice.

Although the risk associated with atopy was substantial, atopy is not a risk factor that can be changed, whereas cumulative mouse allergen exposure can be modified. Furthermore, measures of atopy have low positive predictive values for laboratory animal allergy.²² Lowering cumulative mouse allergen exposure, however, is feasible (whether by changes in caging systems, building design, or use of respiratory protection) and may result in a decreased risk of IgE-mediated mouse sensitization.

Except for rodent/lagomorph sensitivity, specific SPT sensitivities were not significantly associated with mouse SPT sensitivity after controlling for atopy. Because atopy is such

a strong confounder of the relationship between 2 specific allergic sensitizations, and current measures of atopy are suboptimal, it is difficult to know whether the risk associated with rodent/lagomorph sensitization is a result of residual confounding by atopy. If not, this association could also be explained by cross-reactivity of the rodent/lagomorph allergens, by a common exposure history resulting from occupation, or by a genetic predisposition to mount an allergic immune response to this group of animal allergens. Cat or dog SPT sensitivity, which had previously been shown to be a risk factor for rodent SPT sensitivity,²³ was associated with mouse SPT sensitivity but was no longer statistically significant after controlling for atopy. Although these findings suggest an association between some specific SPT sensitivities and mouse SPT sensitivity, prospective studies are needed to refine our current understanding of this association.

Hollander and colleagues⁶ found a dose-response relationship between rat allergen exposure and sensitization using average weekly duration of contact in their exposure metric, but the dose-response relationship was seen only among those who had worked with rats for less than 4 years. The restriction of the finding to those who had shorter duration of employment may be due to healthy worker effect or high-dose tolerance in those working longer than 4 years. Although we cannot completely exclude the possibility that the very highest levels of cumulative exposure were inversely associated with mouse SPT sensitivity, a linear relationship between the log OR of mouse SPT sensitivity and cumulative exposure was a better fit than an inverse "U" relationship, the pattern that would be expected for high-dose tolerance. It could be argued that our study population represents the lower to middle portions of the exposure-response curve and that we did not find evidence to support high-dose tolerance because exposure levels were neither high enough nor long enough. However, a quarter of our study population had worked at The Jackson Laboratory for more than 8 years, and if high-dose tolerance cannot be attained in an occupational setting where some of the highest levels of exposure occur, then it may not be attainable through domestic exposure either.

Although cumulative exposure was related to IgE-mediated mouse sensitization and mIgG and IgG4 levels, current airborne Mus m 1 levels were not. In fact, of the many published studies of rodent allergy, only 1 study⁷ of employees who worked with rats demonstrated that current exposure was related to sensitization in a dose-dependent manner. Our findings suggest that cross-sectional measures of allergen exposure, even airborne measurements, may be poor surrogates for cumulative exposure and could result in biased assessments of the relationship between exposure and sensitization. Although it is true that cross-sectional measures of dust mite and cockroach allergen have been associated with increased risk of IgE-mediated sensitization to those respective allergens,^{24,25} this pattern has generally not held for pet allergens.²⁵ This apparent disparity may simply be a reflection of differences in the immunobiology of these allergens, but it may also be a reflection of the quality of the exposure measure for a given allergen. For example, cross-sectional

measures of cockroach and dust mite allergens may be reasonable surrogates of cumulative exposure because these allergens settle to surfaces and are not easily dispersed.²⁶ More refined exposure measures may be required for animal allergens that tend to be airborne.²⁷

We considered several potential sources of bias in this study. Although there was no association between domestic mouse exposure and SPT sensitivity, home Mus m 1 levels were not measured. To our knowledge, no studies have examined domestic mouse allergen exposure in settings comparable to Maine, where employees of The Jackson Laboratory reside, so the contribution of domestic exposure in this population is unknown. However, if a substantial proportion of total lifetime exposure were attributable to domestic Mus m 1 exposure, then we would have expected the relationship between cumulative occupational Mus m 1 exposure and sensitization to be attenuated. Additional exposure misclassification may have resulted from using duration of employment as a surrogate for duration of exposure. However, more than 85% of the workers had held no more than 2 different jobs, and, of those, 90% maintained a similar level of mouse contact when they changed jobs. Healthy worker bias is always a concern in occupational studies²⁸ and may have affected our results with current exposure. Although rates of job changes in The Jackson Laboratory were similar between the mouse SPT positive and negative groups, the distribution of sensitized workers among current exposure quintiles could still be a reflection of healthy worker bias. However, a healthy worker effect would have decreased the sensitization rate among those in the highest quintiles of cumulative exposure, and we would not see the steady increase in the rate of sensitization shown in Figure 1B.

CONCLUSION

We demonstrated an increasingly monotonic relationship between mIgG and mIgG4 antibody responses and cumulative Mus m 1 exposure. Prevalence rates of IgE-mediated sensitization and elevated levels of mIgG and mIgG4 increased with increasing exposure and were highest among those with the greatest Mus m 1 exposure. These findings provide strong support for ongoing efforts to reduce allergen exposure in occupational settings and suggest that lowering room concentrations of Mus m 1 may prevent or prolong the time to IgE-mediated sensitization. Although we could not definitively exclude high-dose tolerance, we found no evidence to support high-dose tolerance through environmental Mus m 1 exposure.

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