INFLUENCE OF AGE, GENDER, AND ALLERGY STATUS ON NASAL REACTIVITY TO INHALED CHLORINE

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Upper-respiratory-tract symptoms (nasal irritation, congestion and rhinorrhea) are prevalent complaints in so-called “problem buildings.” In epidemiologic studies, symptom reporting is associated with selected subject characteristics, including younger age, female gender, and the presence of allergic rhinitis. The physiologic correlates of these differential reporting patterns, however, are largely unknown. Using dilute chlorine gas as a model upper-respiratory-tract irritant, we studied 52 otherwise healthy volunteers in a sample stratified on age (18–69 yr), gender, and allergy status. In a single-blinded crossover study, subjects had their nasal airway resistance measured preexposure, immediately postexposure, and 15 min postexposure to both filtered air and chlorine (1.0 ppm in air) for 15 min. Allergic rhinitic subjects showed a significantly greater net (chlorine minus air) congestive response at 15 min postexposure than did nonrhinitic controls (p < .01). Advancing age also predicted a greater response immediately post-exposure (p < .01). No gender effect was observed. Significant interindividual variability was evident in the nasal congestive response to irritant (chlorine) provocation. The rhinitis effect was consistent with prior observations, whereas the effect of advancing age was opposite to that hypothesized.

Upper airway irritation and associated reflex endpoints (nasal congestion and rhinorrhea) are commonly reported symptoms in both industrial workplaces and so-called “problem buildings.” Water-soluble/reactive air pollutants and coarse particulate matter disproportionately impact the upper versus lower airway (Dahl, 1990). The resulting upper-respiratory-tract and eye irritation (collectively referred to as “sensory irritation”) acts as a warning to reduce further exposure, in some cases preventing more serious health outcomes (U.S. PHS, 1986).

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Self-reported sensory irritation has been documented in a number of epidemiologic field investigations carried out in buildings with perceived indoor air quality problems. Potential exposures include combustion products (environmental tobacco smoke, reentrained vehicle exhaust, combustion appliances), ozone from photocopying machines, volatile organic compounds or VOCs (cleaning products, office supplies, off-gassing from furniture and building materials, microbial sources), and extremes of temperature and humidity (Hodgson, 2002; Jaakkola et al., 1994; Junker et al., 2001; Molhave, 1992; Ten Brinke et al., 1998). Although environmental sampling has identified specific offending indoor air pollutants in only a minority of cases, consistency has emerged in symptom-reporting patterns, highlighting potential personal risk factors. Females, younger workers, and individuals with preexisting upper airway allergies (rhinitis) are more likely to report symptoms given equivalent exposures (Brasche et al., 2001; Cummings et al., 1991; Hall et al., 1993; Mendell, 1993; Menzies & Bourbeau, 1997; Stenberg & Wall, 1995). By and large, these observed patterns have been interpreted as reflecting psychosocial, rather than biological, differences between subgroups (Brasche et al., 2001; Ryan & Morrow, 1992; Stenberg & Wall, 1995). However, population variability in objective “nasal irritant sensitivity” (sensory acuity and physiologic reactivity) has been incompletely studied (Shusterman, 2002).

Experimentally, the phenomenon of augmented nasal physiologic responsiveness to irritants in allergic rhinitics was first documented by Bascom and colleagues (1991), who measured the nasal congestive response to environmental tobacco smoke (ETS) exposure using posterior rhinomanometry. Although their study group was stratified by self-reported reactivity to ETS, their “historically sensitive” group had substantially higher rates of skin test reactivity to aeroallergens than did the “historically insensitive” group, raising the possibility that allergy status, not self-report, was the relevant biological variable. We have similarly studied the nasal congestive response to irritant provocation using dilute chlorine gas (Cl₂) in otherwise healthy volunteer subjects with and without allergic rhinitis. In a series of counterbalanced, single-blinded, crossover studies, we have found that exposure to Cl₂ (0.5–1.0 ppm × 15 min by nasal mask) produces significant increases in nasal airway resistance (NAR) in seasonal allergic rhinitic (SAR) but not nonrhinitic (NR) subjects (Shusterman et al., 1998, 2002, 2003). As our previous studies were of modest overall size (16–24 subjects) and limited age range (18–40 yr), we have hitherto been unable to examine more than one imputed susceptibility marker. Here, we study a total of 52 subjects spanning the range of 19–68 yr and having relatively symmetrical gender and allergy distributions, with the objective of studying age, gender, and allergic rhinitis status as potential markers of nasal physiologic reactivity to an inhaled irritant.

METHOD

**Subject Recruitment and Screening**

Subjects were recruited through posters and newspaper advertisements. Inclusion criteria were age 18–69 yr and “general good health.” Exclusion
criteria were: (1) a history of asthma, (2) cigarette smoking (active or within previous 6 mo), (3) pregnancy or lactation, (4) a history of severe allergic reactions (anaphylaxis or angioedema), and (5) continuous therapy with medications having antihistaminic side effects (e.g., tricyclic antidepressants). Subjects read and signed an informed consent document approved by the Committee on Human Research of the University of California, San Francisco. Questionnaires were administered to each potential subject, who was provisionally classified as having seasonal allergic rhinitis (SAR), no rhinitis (NR), or “other” (including perennial allergic rhinitis).

Allergy skin-prick tests (to 13 regionally common aeroallergens/mixes, plus saline and histamine controls) were then administered. For purposes of this study, “seasonal allergic rhinitics” were defined as subjects with: (1) a history of seasonally occurring sneezing, nasal pruritus, rhinorrhea, postnasal drip, and/or nasal congestion, with or without known precipitants; and (2) skin-test reactivity to at least one seasonally occurring agent from the panel that corroborated the history. “Skin-test reactivity” was defined as a wheal reaction to skin-prick testing with a diameter greater than or equal to that of the histamine control. “Nonrhinitics” were defined as subjects who report, at most, infrequent nasal symptoms, without identified seasonal variation or precipitants, with significant skin test reactivity to no more than 1 agent in the panel of 13 aeroallergens, and with normal findings on anterior rhinoscopy. Prior to skin testing, subjects were asked to refrain from taking antihistamines for 72 h (hydroxyzine for 3 wk, astemizole for 12 wk).

All potential subjects for this study first participated in a study of sensory thresholds to pulsed carbon dioxide (detection) and to vapor-phase n-propanol (localization; Shusterman et al., in press). During the course of that study, subjects were screened for their ability to generate meaningful tracings by active posterior rhinomanometry, and 8 of 60 subjects were unable to do so, despite extensive coaching. The two study sequences were separated by at least 2 wk, and as the sensory measures were of a threshold nature, subjects experienced minimal subjective irritation and there was minimal potential for stimulus carryover or “priming.”

**Experimental Design and Procedures**

The study design was experimental, utilizing a semirandomized crossover design comparing the effect of dilute Cl₂ gas with that of air (Figure 1). The concentration and duration of Cl₂ exposure—1.0 ppm times 15 min—is the (U.S.) occupational short-term exposure limit, and hence is of both scientific and regulatory interest. The two exposures (a week apart) employed active posterior rhinomanometry, with nasal airway resistance measured at baseline, immediately after exposure and again 15 min postexposure. On a given day, exposure was either to pure (medical grade) air or to Cl₂ (1.0 ppm) diluted in air. The order of exposure within each pair of testing dates was determined by limited randomization (i.e., with initial exposure for subjects in each gender/rhinitis/age group stratum alternating between air and Cl₂). For stratification purposes, subject age was divided into three groups: 18–34, 35–51, and 52–69 yr.
SAR subjects were tested out of their relevant pollen season. All subjects were asked to avoid exercising, consumption of spicy foods, and use of scented cosmetics on the day of testing. In addition to antihistamine preclu-
sions (as specified for skin-prick testing), subjects were asked to avoid using nasal steroids for at least 2 wk, and nasal decongestants for at least 48 h prior to testing. Upon arrival at the laboratory, subjects entered a climate-
controlled chamber (22 ± 1°C; 40 ± 3% relative humidity [RH]) with filtered air (activated charcoal and high-efficiency particulate). After a 15-min wait-
ing period, baseline symptoms (nasal irritation, congestion, rhinorrhea, post-
nasal drip, and odor) were rated on computer-based visual analog scales or 
VAS (LabView software, National Instruments, Austin, TX). The scales were 
indexed at equal intervals with the words “none,” “slight,” “moderate,” 
“strong,” “very strong,” and “overpowering,” corresponding to the numerical 
range of 0.00 to 5.00. Symptom rating was repeated (times three) at 5-min intervals during exposure, and again at 15 min postexposure.

Cl₂ or air was administered on a single-blinded basis for a period of 15 
min. Cl₂ was diluted in medical-grade air, which was preconditioned to 22°C 
and 40% RH (verified using the same temperature/humidity recorder em-
ploved in the chamber itself); the dilution apparatus has previously been de-
scribed in detail (Shusterman et al., 1998). Cl₂ and air were supplied by Puri-
tan Medical Products (Hayward, CA). Cl₂ concentrations were measured in 
real time using an electrochemical monitor (model 1340; Interscan Corp., 
Chatsworth, CA). The meter was recalibrated on a daily basis using the cer-
tified contents of the Cl₂ cylinder as the standard. The gas mixture was administered to the subject through a nasal CPAP mask (series 3121; Respironics, Inc., Murraysville, PA), which was sized according to the individual subject. The combination of a high flow rate (60 L/min) and a low-pressure gas scavenging system allowed subjects to breathe with negligible superimposed pressure or resistance.

Nasal airway resistance for each testing condition was taken as the mean of three values, as ascertained by active posterior rhinomanometry using a commercial instrument (model NR6-2, GM Instruments, Kilwinnig, UK). NAR was calculated using the pressure-cutoff method (75 Pa), and was obtained at baseline, immediately postexposure, and 15 min postexposure on both Cl₂ and air days. Calibration procedures were employed as previously detailed (Shusterman et al., 1998).

Data Analysis

NAR values were normalized to baseline measurements for each experimental session, yielding the metric “Δ% NAR” [from baseline] for each of two time points (immediately postexposure and 15 min postexposure) on each of two (Cl₂ and air) experimental conditions. The difference between the 2 days was further summarized as a net measure (Cl₂ minus air day): “Δ% NAR (Net).” Data were examined for normality and then analyzed by analysis of variance (ANOVA) if this condition was satisfied. The hypotheses to be tested were: (1) SAR subjects would show significantly greater Cl₂-related increases in NAR over baseline [“Δ% NAR (Net)’’] than would NR controls, (2) younger subjects would show significantly greater increases in NAR than older subjects, and (3) females would show significantly greater increases in NAR than males.

RESULTS

The demographic and health characteristics of the 52 participants are summarized in Table 1. Subject ages ranged from 19 to 68 yr, with 24 males (46%) and 27 allergic rhinitics (52%). Seventeen subjects were recruited in the 18–34 yr age range, 18 in the 35–51 yr range, and 17 in the 52–69 yr range. Overall, 15 of 28 females were allergic rhinitic (54%) compared with 12 of 24 males (50%). The mean ages of males and females were similar, as

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>Status (number)</th>
<th>Age: mean ± SD (yr)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gender</td>
<td>Male (24)</td>
<td>40.9 ± 14.8</td>
</tr>
<tr>
<td></td>
<td>Female (28)</td>
<td>41.8 ± 14.2</td>
</tr>
<tr>
<td>Allergic rhinitis</td>
<td>Yes (27)</td>
<td>40.9 ± 13.6</td>
</tr>
<tr>
<td></td>
<td>No (25)</td>
<td>41.8 ± 15.3</td>
</tr>
<tr>
<td>Combined</td>
<td>(52)</td>
<td>41.3 ± 14.3</td>
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</table>
were the mean ages of allergic rhinitics and nonrhinitics (Table 1; \( p > .80 \)). Given this nearly balanced study sample, study power could be maximized in subsequent analysis by utilizing univariate models.

The exposure metric, \( \Delta % \text{NAR (Net)} \), was normally distributed, both immediately postexposure and 15 min postexposure, and thus was analyzed by ANOVA. Results by age, gender, and rhinitis status, both immediately postexposure and 15 min postexposure, appear in Figure 2. At 15 min postexposure, SAR subjects showed a significantly greater congestive response to \( \text{Cl}_2 \) (compared to air) than did nonallergic controls \( (p < .01) \). An enhanced response to \( \text{Cl}_2 \) was also seen as a function of age, although the effect was opposite of that predicted (i.e., older subjects congested more than younger; \( p < .01 \) immediately postexposure). Gender did not significantly predict the congestive response to \( \text{Cl}_2 \) at either sampling time, although there was a non-significant trend \( (p = .15) \) toward greater \( \Delta % \text{NAR (Net)} \) in males than females at 15 min postexposure.

Subjectively rated nasal irritation, congestion, rhinorrhea, postnasal drip, and odor were all quite modest in this experiment. The mean peak VAS ratings for odor and irritation during exposure, for example, were less than “slight” (i.e., <1 on a scale of 0–5), and showed the predicted time course of response of adaptation for odor and temporal integration for irritation (Figure 3). There was no significant differential symptom rating by rhinitis status, age, or gender, and the remaining symptoms (congestion, rhinorrhea, postnasal drip) were of even lower magnitude (data not shown).

![FIGURE 2. \( \Delta % \text{NAR (Net)} \) measured immediately (Post-1) and 15 min (Post-2) after (\text{Cl}_2 \) minus air) exposure, as a function of (a) rhinitis status, (b) age, and (c) gender. \( \Delta % \text{NAR (Net)} \) increased significantly with age immediately postexposure \( (p < .01) \) and was significantly greater in allergic rhinitics than in nonrhinitics 15 min postexposure \( (p < .01) \). The difference between males and females at 15 min postexposure was not statistically significant \( (p = .15) \).]
DISCUSSION AND CONCLUSIONS

In this sample, we found that allergic rhinitis predicts a greater congestive response to irritant provocation (i.e., increases in NAR measured at 15 min postexposure), similar to findings in our earlier studies (Shusterman et al., 1998, 2002, 2003). In addition, we found that more advanced age predicted a more marked congestive response, as measured immediately postexposure. Finally, gender did not emerge as a significant predictor of physiologic re-
activity in this study sample. The observed changes in NAR occurred with very modest subjective nasal irritation.

This study represents an extension of our previous descriptive work on Cl₂-induced nasal congestion, with the upper age limit for recruitment being extended from 40 to 69 yr, and with an overall study size (52) sufficiently
large to examine more than one susceptibility marker. By employing targeted recruiting into age/gender/allergy strata, we were able to construct a sufficiently balanced study sample to permit optimization of statistical power using univariate statistical methods (i.e., ANOVA). Further, order of exposure was counterbalanced within strata, effectively eliminating stimulus order or stimulus carryover as potential confounders. Although “single blinding” of exposure is only partially effective with an odorous air pollutant, suprathreshold ratings of both odor and irritation were quite modest, and roughly one of four subjects were unable to correctly identify Cl₂ versus air exposure days.

Of our two significant physiologic findings, the positive correlation between age and congestive response to Cl₂ was contrary to that predicted by epidemiologic reporting patterns. This finding is surprising in light of the clinical assumption that rates of “atrophic rhinitis” increase with age and that this condition is accompanied by impaired vascular reactivity (Bende, 1985). Further, these findings may signal a “disconnect” between sensory and physiologic data. Specifically, in a largely overlapping cohort of 60 subjects, we found that older age predicted higher irritant detection thresholds (i.e., less “sensitivity”), utilizing carbon dioxide and n-propanol as test compounds (Shusterman et al., in press). Another difference between sensory and physiologic endpoints in these two studies included the finding of significantly lower irritant (CO₂) detection thresholds in female subjects, but no gender difference in the congestive response to Cl₂.

The mechanism(s) involved in the nasal congestive response to irritant provocation are unclear. Despite the link between physiologic reactivity to irritants and nasal allergy status, mast-cell degranulation seems unlikely in light of nasal lavage findings after provocation with both environmental tobacco smoke (Bascom et al., 1991) and Cl₂ (Shusterman et al., 2003). Further, cholinergic parasympathetic reflexes seem to have variable role in this phenomenon, based on studies in which cholinergic blockers were administered prior to provocation with ammonia (McLean et al., 1979) versus Cl₂ (Shusterman et al., 2002). Interest has been expressed in the potential involvement of vasoactive neuropeptides released by sensory nerves (Bascom, 1992). However, a recent study examining the congestive response to a hyperosmolar irritant stimulus found evidence for neither mast-cell degranulation nor neuropeptide (substance P) release (Koskela et al., 2000). Should airway sensory nerves prove to be involved in irritant-induced nasal congestion, however, the logical differential response mechanism for allergic rhinitics would be so-called “neuromodulation” (Undem et al., 2000).

Notwithstanding ambiguities surrounding the mechanism(s) of irritant-related nasal congestion, the finding of differential nasal responsiveness to irritant air pollutants is one that should be kept in mind by clinicians, risk assessors, and public health practitioners. The common practice of ascribing differential upper-airway symptom reporting to psychosocial factors alone is one that should be tempered in light of human experimental data demonstrating heterogeneity of response in objective tests of sensory acuity and physiologic reactivity. Studies such as these may have further value by allowing substitution of empirical data for default assumptions regarding interindi-
Individual variability when conducting risk assessments including the upper-airway impact of irritant chemicals.

REFERENCES


