Original Articles

Increase in non-specific bronchial hyperresponsiveness without specific response to isocyanate in isocyanate-induced asthma: a pilot study

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Increased non-specific bronchial hyperresponsiveness (BHR) has been reported after positive reaction to isocyanates in patients with isocyanate-sensitive asthma. The increased responsiveness may, however, also precede the asthma attack. We therefore compared non-specific BHR to a cholinergic agent before and after exposure to toluene-diisocyanate (TDI) that induced no asthma symptoms in 11 workers with isocyanate-related asthma. Patients were exposed for 3 consecutive days to progressively increasing doses of TDI (5, 10, and 20 ppb min^-1 for 20 min) in an exposure chamber with continuous TDI monitoring. No immediate nor late asthmatic bronchial reaction was observed in any patient after any dose of TDI during or after challenge. A significant increase in non-specific BHR was noted 24 h after the last dose of TDI challenge, however. This increase was at least one doubling dose for seven of 11 patients.

In conclusion, our study shows that, in patients with isocyanate-induced asthma, exposure to TDI induces a slight but significant increase in non-specific BHR in the absence of any immediate or late bronchial response to isocyanate. This result, which requires further confirmation, may justify a proposal to measure non-specific BHR, even after a negative specific inhalation test to TDI, as an additional diagnostic element for TDI-induced occupational asthma, to help lower the percentage of the undetected occupational asthma cases.

Introduction

Increased bronchial hyperresponsiveness (BHR) after a positive allergen challenge test has been widely documented (1–3). Similarly, BHR increases after a positive bronchial provocation test to toluene diisocyanate (TDI), both after late or dual reactions (4) and after an immediate response (5). In allergic asthma, repeated inhalation of low doses of allergens that do not provoke by themselves any bronchoconstrictive response may induce an increase in non-specific BHR (6). Hence, we hypothesized that non-specific BHR might also increase after a negative occupational allergen challenge and might be useful in diagnosing occupational asthma. This aspect of BHR has hitherto received little attention (7,8).

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We report here the changes in non-specific BHR in 11 patients with isocyanate-induced asthma who were exposed to increasing low doses of isocyanate that did not induce a positive bronchial reaction.

Patients and Methods

PATIENTS

This study included 11 patients: 10 men and one woman, aged 27–46 years (mean: 36.1 ± 7.3 years) (Table 1). All had a clinical history of isocyanate-induced asthma, characterized by chest tightness, dyspnoea, and wheezing on work days which was reversible with β2-agonists. Symptoms improved or disappeared on days off (9). All patients were hospitalized for the study. These patients were chosen because they had no positive reaction to low doses of TDI administered on 3 consecutive days (5, 10, 20 ppb).

Five patients (nos 1, 2, 4, 6, 11) had completed a 1-month diary card that listed their workdays, tasks, and symptoms
TABLE 1. Characteristics of the patients before and after TDI challenge

<table>
<thead>
<tr>
<th>Patient</th>
<th>Age</th>
<th>Before TDI FEViB (l)</th>
<th>After TDI FEViB (l)</th>
<th>ΔFEViB (%)</th>
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<tbody>
<tr>
<td>1</td>
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<td>3.5</td>
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<td>3</td>
<td>41</td>
<td>2.4</td>
<td>2.5</td>
<td>4.2</td>
</tr>
<tr>
<td>4</td>
<td>31</td>
<td>3.0</td>
<td>2.9</td>
<td>3.3</td>
</tr>
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<td>5</td>
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<td>4.3</td>
<td>4.3</td>
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<td>2.8</td>
<td>2.7</td>
<td>2.9</td>
</tr>
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<td>39</td>
<td>3.6</td>
<td>3.6</td>
<td>1.4</td>
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<td>9</td>
<td>41</td>
<td>3.8</td>
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<td>45</td>
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<td>3.5</td>
<td>2.8</td>
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<td>11</td>
<td>27</td>
<td>3.3</td>
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</tr>
<tr>
<td>Mean</td>
<td>36.1</td>
<td>3.4</td>
<td>3.4</td>
<td>2.5</td>
</tr>
<tr>
<td>SD</td>
<td>7.3</td>
<td>0.5</td>
<td>0.5</td>
<td>1.4</td>
</tr>
</tbody>
</table>

FEViB, baseline FEVi before cholinergic challenge.

for the month. Peak flow was evaluated at least three times daily during that month. Changes between the maximum value obtained on days off and the minimal PEF value observed at the workplace were calculated as the percentage: (max−min)/max × 100. Peak flow values of all five patients dropped significantly (>20%) on work days.

Two of 11 patients had specific IgE to TDI. Six of 11 patients had positive skin prick tests to indoor allergens, pollens, or both, and only one had asthma before exposure to TDI (horse and dog contact). Two patients were smokers and six were ex-smokers.

Patients were not exposed to isocyanates at work for at least 2 days before inclusion and were hospitalized during the study for extensive investigation. All medications were stopped before the study: 15 days before for inhaled corticosteroids and 12 h before for immediate β2-agonists.

STUDY DESIGN

Patients underwent a cholinergic provocation test to evaluate bronchial responsiveness 1–4 days before the initial TDI challenge. They were then challenged on 3 consecutive days with TDI at increasing doses (5, 10, and 20 ppb for 20 min on days 1, 2, and 3, respectively). A second cholinergic provocation test was performed 24 h after the highest TDI dose (20 ppb), and the two dose–response curves were compared.

METHODS

Provocation tests to the cholinergic agent

The non-specific bronchoprovocation test was carried out by administering increasing doubling doses of the cholinergic agent. Methacholine aerosol (solution 1/100 in saline) was generated from a starting volume of 2 ml in a disposable Minineb 5610 5610 DeVilbiss nebulizer (Somerset, PA, U.S.A.) by means of a breath-activated dosimeter (FDC 88; Mediprom, Paris, France) under a pressure of 1.5 bar. During inhalation through a mouthpiece, the subject wore a noseclip. Each dose of aerosol was delivered during a breath taken slowly with a 2–5 s breath-hold from residual capacity to near total lung capacity. The nebulizer delivered 4, 8 or 16 μl per breath depending on the nebulization time (0.6, 1.2 or 2.4 s), leading to inhalation of cumulative doubling doses of 25–3200 μg methacholine. Doses were expressed in base 2 logarithm, i.e. log2 doses from 1 to 8.

Airway responses were assessed by the measurement of the FEVi with an automated flowmeter (Autospiro AS500, Minato, Osaka, Japan). Challenges were preceded by the inhalation of 0.9% NaCl, which caused no or less than 5% change in FEVi in any of the subjects. A positive test was stopped when a fall in 20% in baseline FEVi occurred. Two puffs of β2-agonist were administered and the reversibility of the bronchospasm was evaluated by measuring FEVi 10 min later.

When a fall of less than 20% was observed for the highest dose of the cholinergic agent, the test was considered negative.

TDI provocation test

The inhalation of TDI took place in a dynamic flow chamber where the airborne concentration of isocyanate was continuously measured with an MDA monitor (1/100 MDA Scientific, Grenview, IL, U.S.A.). Exposure lasted for 20 min.

TDI was generated by heating 10 ml of TDI in a closed cell, with a controlled flow rate into a mixing chamber. There, a fan ensured adequate mixing and circulation of air with the TDI vapour. FEVi was measured before exposure, immediately after, and hourly for 8 h. If symptoms occurred later, FEVi was measured again. For this reason, the patients stayed in the hospital for 3 days.

EXPRESSION OF THE RESULTS AND STATISTICAL ANALYSIS

Individual dose–response curves were constructed and PD20, i.e. the dose inducing at least a 20% fall in FEVi, was calculated for each subject. Doubling doses were used for both tests, in accordance with previous studies of between-test reproducibility of cholinergic challenge tests with the dosimeter method (10).

Two-way analysis of variance (ANOVA) was used to analyse the curves (Statview II, Abacus concept, U.S.A.).

To test for changes in cholinergic airway responsiveness, the logarithm base 2 values for cholinergic doses (PD20) before and after TDI inhalation were compared by a Wilcoxon rank test. When the cholinergic test before TDI challenge was negative at the highest dose used (3200 μg MCh), the positive dose was considered as the dose
immediately above, which corresponds to the less favourable hypothesis. A $P$ value lower than 0.05 was considered significant.

Results

**TDI INHALATION**

All 11 subjects tolerated the inhalation of TDI up to 20 ppb without either an immediate or a late asthmatic reaction.

**CHALLENGE TEST TO THE CHOLINERGIC AGENT**

The baseline FEV$_1$ of each subject was identical on both study days (Table 1, Fig. 1).

Specific BHR to the cholinergic agent before the TDI test was negative in three patients (nos 1, 6, 10). One of them (no. 1) showed non-specific BHR 24 h after the last TDI challenge, with a variation of one doubling dose before and after TDI (Table 2, Fig. 1). Of the eight subjects with positive cholinergic provocation tests before TDI challenge, airway responsiveness increased in six (nos 2, 3, 5, 8, 9, 11) 24 h after the last dose of TDI. Two patients exhibited a change of three doubling doses before and after TDI (no. 3, 9), and four patients had a change of one doubling dose (Table 2). In the two remaining patients (nos 4, 7), airway responsiveness to the cholinergic agent did not vary.

For the group as a whole, the dose–response curves to the cholinergic agent before and after TDI administration of TDI differed significantly ($P<0.01$) (Fig. 1). PD$_{20}$, i.e. the provocation doses inducing at least a 20% fall in FEV$_1$, decreased significantly ($P<0.01$) by a doubling dose in four patients and by more than a doubling dose in three others (Fig. 1).

In the nine patients with positive cholinergic provocation after TDI challenge, mean FEV$_1$ fall at PD$_{20}$ was 23.5% after challenge; before the challenge it was 11.8% at the same dose of methacholine ($P<0.02$) (Table 2).

Discussion

Our results show that a TDI provocation challenge that induced no fall in FEV$_1$ in isocyanate-sensitive patients led to a slight but significant increase in non-specific BHR. This increase may be related to the observed increase in non-specific BHR after exposure of workers to their work environment (11–14).

One strength of our study is that optimal study conditions. The patients were hospitalized, i.e. away from work and isocyanates. They were also clinically stable. Mean baseline FEV$_1$ varied by less than 3%. These excellent conditions resulted in a 95% confidence interval for PD$_{20}$ of less than the difference of one doubling dose (15).

Increases occurred after patients were exposed to permissible exposure levels of isocyanates, between 5 ppb and the threshold value of 20 ppb. In our group of 11 patients, three had negative cholinergic provocation tests before exposure to the specific inhalation challenge test. This result conforms with observations in larger series of TDI-asthmatic patients, which report that between 10% and 21% of patients had negative methacholine tests (16–18).

Several studies report that a positive TDI challenge test in TDI-sensitive asthma is followed by an increase in BHR to a cholinergic agent. This result has been observed most often after a late reaction to TDI (4,12), but also after immediate isolated reactions (5). The temporal relationship between increased non-specific BHR and late asthmatic responses induced by several occupational agents, including TDI, was studied by Durham et al. (13), who found that the increase in histamine responsiveness takes place approximately 3 h after exposure to TDI, thus preceding the late asthmatic response. BHR remained elevated 24 h after TDI challenge, although its intensity was lower. In our study, even in the absence of a positive TDI challenge test, a significant, although slight, increase in BHR occurred 24 h after challenge. Since the patients were followed clinically for 24 h with repeated functional respiration tests after the last TDI inhalation, any positive early or late specific response would have been detected. It is therefore very unlikely that the observed increase in BHR was related to a late response. Hence, an increase in BHR may precede a positive TDI response. Several other results support this conclusion. These include a case study reporting an increase in non-specific BHR without any positive response to red cedar allergen (15), and the study by Ihre et al. (6), in which repeated inhalations of low doses of Aeroallergens increased non-specific BHR in 11 of 13 patients with seasonal allergic asthma. Finally, this conclusion is consistent with experimental studies of occupational disease performed in animals: non-specific BHR increases after exposure to low doses of TDI in the absence of a specific response (19).

Our results might be important in lowering the percentage of undetected occupational asthma cases, which is suspected to be high. Various studies report that, of patients who have respiratory symptoms and are exposed to isocyanate in the workplace, only half have positive isocyanate tests (18,20). Proposed explanations have included the timing of the challenge test (at a moment when the patients had been unexposed for a long period) and a lower duration and doses of laboratory exposure, which, because limited to the 20 ppb threshold dose of TDI for 10 min, were lower than those inducing asthma symptoms in the workplace. One conclusion to be drawn from our finding that BHR increased although there was no positive TDI reaction is that the patients were sensitive to TDI. Measuring BHR after provocation with low doses of TDI might therefore decrease the number of cases of TDI-induced asthma that go undetected. Support for this comes from a recent report that patients with a modified BHR 6 h after a first negative challenge test to an occupational agent reacted positively to this agent after a second exposure of 2–3 h (8). This conclusion is also in accordance with our previous finding of a positive TDI-challenge test to 20 ppb occurring after an increase in BHR observed after a 5 and 10 ppb inhalation challenge without positive response (7). Thus, these reports suggest that the performance of
FIG. 1. Individual dose–response curves to the cholinergic agent (methacholine) (ChA, increasing doubling doses) in patients with a clinical history of TDI asthma before (○) and after (●) inhalation of low doses of TDI (5, 10 and 20 ppb) on 3 consecutive days. The FEV₁ theoretical values (%) are represented as a function of the dose (log₂). b=FEV₁ at the beginning of the test. p=FEV₁ after inhalation of placebo (saline).
TABLE 2. Cholinergic provocation lasts before and after TDI challenge

<table>
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<tr>
<th>Patient</th>
<th>PD 20 (log 2)</th>
<th>% fall FEV1</th>
<th>% fall observed after TDI</th>
<th>PD 20 (log 2)</th>
<th>% fall FEV1</th>
<th>% fall FEV1 at PD 20</th>
<th>Δ log PD 20</th>
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<tr>
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<td>11.8†</td>
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<td>SD</td>
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*: for patient 1, since a fall in FEV1 of less than 20% occurred at dose 8, a 20% fall in FEV1 was extrapolated as dose 9.
†: p<0.02 between before and after TDI for % fall FEV1 at PD 20 observed after TDI at cholinergic challenges (Wilcoxon paired rank test).

In conclusion, our study shows the occurrence of a slight but significant increase in BHR after patients with a clinical history of asthma are exposed to repeated low doses of isocyanate. This result may be important in view of the possibly high percentage of undetected cases of occupational asthma. These results, which require confirmation, may justify a proposal to measure non-specific BHR, even after a negative specific inhalation test to TDI, as an additional diagnostic element for TDI-induced occupational asthma.

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References


