

Is occupational asthma to diisocyanates a non-IgE-mediated disease?

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Background: Exposure to diisocyanates in the workplace is an important cause of occupational asthma. The majority of patients with diisocyanate-induced asthma have no detectable diisocyanate-specific IgE antibodies in serum. There has been much debate as to whether this is due to diisocyanate-induced asthma being mediated by non-IgE mechanisms or whether it is the result of using inappropriate conjugates. **Objective:** We sought to determine whether RNA message for C ϵ , IL-4, and other associated inflammatory markers could be detected locally within the bronchial mucosa after diisocyanate challenge.

Methods: Fiberoptic bronchoscopic bronchial biopsy specimens were obtained at 24 hours after both a control and an active challenge in 5 patients with positive and 7 patients with negative inhalation test responses to diisocyanates. Using both immunohistochemistry and *in situ* hybridization, we determined mRNA for C ϵ , IL-4, IL-5, and other associated inflammatory markers.

Results: There was a striking absence of C ϵ and IL-4 mRNA-positive cells in bronchial biopsy specimens from patients challenged with diisocyanate (C ϵ median of 0 and interquartile range of 0-1.85; IL-4 median of 0 and interquartile range of 0-0.85). In contrast, there were increased numbers of IL-5-, CD25-, and CD4-positive cells and a trend toward an increase in eosinophils after active challenge with diisocyanate.

Conclusion: We found a striking absence of both bronchial C ϵ and IL-4 RNA message after inhalation challenge with diisocyanates, irrespective of whether the challenge test response was positive or negative. We propose that diisocyanate-induced asthma is a non-IgE-mediated disease, at least in patients in whom specific IgE antibodies to diisocyanates are undetectable. (*J Allergy Clin Immunol* 2006;117:663-9.)

Key words: Occupational asthma, isocyanates, IgE

Diisocyanates are highly reactive, low-molecular-weight chemicals with widespread industrial applications.

Abbreviations used

HDI: Hexamethylene diisocyanate
IQR: Interquartile range
MDI: Diphenyl-methyl diisocyanate
TBS: Tris-buffered saline
TDI: Toluene diisocyanate

Toluene diisocyanate (TDI), diphenyl-methyl diisocyanate (MDI), and hexamethylene diisocyanate (HDI), in particular, are used to produce a variety of polyurethane-based products. Exposure to diisocyanates in the workplace is an important cause of occupational asthma, inducing disease in between 5% and 15% of exposed employees.¹⁻⁴ An estimated 20% of cases of occupational asthma in the United Kingdom⁵ and 50% in Ontario (before the introduction of control measures)⁶ are attributed to diisocyanate exposures.

For many years, it has been recognized that only a minority of cases of diisocyanate-induced asthma are accompanied by detectable levels of serum-specific IgE antibody.⁷ In case series in which the diagnosis has been confirmed by means of specific inhalation testing, this proportion has been between 14% and 28%.⁷⁻¹⁰ Diisocyanates are haptens and require conjugation to a carrier protein for the detection of specific IgE antibodies. Contemporary methods for generating diisocyanate-albumin conjugates have changed little since they were first described, and they remain poorly standardized. It is a matter of debate to what extent specific IgE antibody production is not detected because of inappropriate conjugates¹¹ and to what extent the disease is mediated by alternative immunologic or nonimmunologic mechanisms.

Significantly increased levels of C ϵ and IL-4 message are detected in the bronchial mucosa after active challenge in patients with asthma caused by high-molecular-weight allergens, such as grass pollen or house dust mite, and also in those with apparently intrinsic disease.¹² The production of IgE requires both IL-4 to target the ϵ gene for recombination and cross-linking of CD40 on the B cell with CD40 ligand on T lymphocytes or basophils to allow isotype switching to take place.¹³ The somatic recombination of the immunoglobulin heavy chain cluster gives rise to mRNA for the ϵ heavy chain. In this study we set out to examine whether C ϵ , IL-4, and other associated inflammatory markers could be detected locally within the bronchial mucosa after diisocyanate challenge.

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TABLE I. Clinical summary of patients with diisocyanate-induced asthma

Agent	Age (y)	Occupation	Total IgE (kU/L)	Specific IgE* (kU/L)	Atopic	Duration away from exposure (mo)	Maximum % change in FEV ₁	PC ₂₀		Challenge result
								Control	Active	
HDI	38	Paint sprayer	392.2	<0.35 <0.35 <0.35	Y	48	−28%	0.18	0.15	+
HDI	35	Paint sprayer	16.2	<0.35 <0.35 <0.35	Y	1	−32%	>16	5.8	+
HDI	21	Paint sprayer	116	<0.35 <0.35 <0.35	Y	1	−60%	4.7	4.2	+
HDI	38	Injection molder	22.2	<0.35 <0.35 <0.35	Y	<1	−40%	0.3	0.16	+
HDI	28	Paint sprayer	110.5	<0.35 <0.35 0.5	N	3	−20%	>16	>16	+
HDI	27	Injection molder	32.2	<0.35 <0.35 3.5	Y	5	0	>16	13	−
HDI	39	Health and safety	79.9	<0.35 <0.35 <0.35	Y	2	0	3.6	2.7	−
MDI	31	Molding technician	274.8	<0.35 <0.35 <0.35	Y	<1	0	>16	>16	−
HDI	35	Paint sprayer	118.9	<0.35 <0.35 <0.35	Y	<1	0	>16	>16	−
HDI	38	Paint sprayer	27.2	<0.35 <0.35 <0.35	N	<1	0	6.2	>16	−
HDI	35	Paint sprayer	4.73	<0.35 <0.35 <0.35	N	6	0	>16	>16	−
HDI	41	Paint sprayer	329.4	<0.35 <0.35 <0.35	N	11	0	2.8	3.0	−
Control (n = 6)	31.5		46.0	ND	N		0	>32		ND

ND, Not done.

*HDI, MDI, and TDI, respectively.

METHODS

Bronchoscopy study design

Patients were recruited from a specialist clinic after referral by their general practitioner or occupational health physician. All patients with suspected diisocyanate-induced asthma in whom there was a clinical indication for specific inhalation testing were invited to take part. In this way we recruited 12 patients (all male) between 1997 and 2003; their clinical details are summarized in Table I, and duration of exposure before onset of symptoms and duration of symptoms before leaving work are summarized in Table II.

The study was approved by the Ethics Committee of Royal Brompton and Harefield Hospital, and each patient provided informed consent.

Occupational inhalational testing

Single-blind, controlled inhalation tests were undertaken in an exposure chamber under medical supervision designed to reproduce, as far as practicable, appropriate workplace exposures. Carefully regulated active or control (inactive) exposures were arranged on consecutive days until a diagnosis was reached. Nine patients worked as spray painters and were exposed to an HDI hardening agent, which was omitted from the paint mixture during control challenges. Three patients worked in injection molding, 2 exposed to HDI and 1 to MDI; workplace conditions for the former were reproduced by squirting air through a mixture of the active and inert components, with the active component being omitted during control challenges. MDI exposure was reproduced by heating crystals of the diisocyanate in a sand bath for 15 minutes. Both active and control challenges were monitored in every case by using continuous measurement of airborne diisocyanate concentrations, these being maintained within the appropriate short-term exposure limit values. Responses to each day's challenge were assessed by FEV₁ measurements at 0, 5, 10, 15, and 30 minutes after the exposure and hourly thereafter for up to 12 hours. In addition, nonspecific bronchial responsiveness to histamine was measured at admission and 24 hours after each exposure, apart from the days when bronchoscopy was undertaken.¹⁴ A reproducible and sustained dual or late-phase decrease in FEV₁ of greater than 15% from baseline value, an increase in nonspecific bronchial reactivity after active challenge alone, or both was considered a positive response.

Fiberoptic bronchoscopy

All but 2 patients underwent fiberoptic bronchoscopy 24 hours after a control exposure and again 24 hours after an active exposure. Two patients had one bronchoscopy only, one after a control challenge and the other after an active challenge. On bronchoscopy days, histamine response testing was omitted from the usual schedule. Subjects were premedicated with nebulized salbutamol (2.5 mg), intravenous atropine (0.6 mg), and midazolam (5–10 mg) 10 minutes before the procedure. Oxygen was delivered throughout through nasal cannulae, with oxygen saturation monitored by means of pulse oximetry. The nose and throat were anesthetized with topical lignocaine spray and 4% lignocaine applied through the bronchoscope channel to the vocal cords, with 2% lignocaine used to anesthetize the upper airways. We used an Olympus model BF P20 bronchoscope (Olympus Company, Tokyo, Japan) to obtain 4 to 8 biopsy specimens from segmental divisions of the major bronchi, with bronchoalveolar lavage carried out just before the taking of biopsy specimens. All bronchoscopies and procedures were carried out by the same experienced operator.

Skin prick testing

Skin prick tests were undertaken with physiologic saline solution, histamine, grass pollen, cat, and *Dermatophagoides pteronyssinus* (Allergopharma, Reinbeck, Germany). Wheal sizes were recorded after 10 minutes by measuring the mean of their longest and midpoint orthogonal internal diameters; a positive response was one with a mean wheal diameter of at least 3 mm greater than that obtained in response to saline solution. Atopy was defined as a positive response to tests with one or more extracts of cat, grass pollen, or *D pteronyssinus*.

Specific IgE antibodies

Specific IgE antibodies to diisocyanates were measured with Unicap (Sweden Diagnostics, Milton Keynes, United Kingdom). In only 2 patients were measurements greater than 0.35 kU/L (Table I).

Biopsy specimens

Biopsy specimens were divided into 2 halves. One half was immediately mounted in OCT compound (VWR; Lutterworth, Leics, United Kingdom), snap-frozen by means of immersion in

TABLE II. Duration of exposure and symptoms in patients with diisocyanate-induced asthma

Agent	Age (y)	Occupation	Duration of exposure before onset of symptoms (mo)	Duration of exposure before leaving of work (mo)	Challenge result
HDI	38	Paint sprayer	24	204	+
HDI	35	Paint sprayer	145	23	+
HDI	21	Paint sprayer	34	2	+
HDI	38	Injection molder	5	17	+
HDI	28	Paint sprayer	138	3	+
HDI	27	Injection molder	24	5	—
HDI	39	Health and safety	168	2	—
MDI	31	Molding technician	30	108	—
HDI	35	Paint sprayer	192	24	—
HDI	38	Paint sprayer	107	13	—
HDI	35	Paint sprayer	210	15	—
HDI	41	Paint sprayer	95	2	—

isopentane precooled in liquid nitrogen, and then stored at -80°C . Immunohistochemistry was performed on 6- μm cryostat sections fixed in acetone/methanol (60:40).

The other half was fixed in 4% paraformaldehyde/PBS solution for 2 hours, washed in 15% sucrose/PBS twice, embedded in OCT compound (Merck Ltd, Leicestershire, United Kingdom), and snap-frozen in isopentane precooled in liquid nitrogen. *In situ* hybridization was performed on 6- μm sections cut on poly L-lysine microscope slides (Merck), dried overnight at 37°C , and stored at -80°C .¹⁵

Immunohistochemistry

Monoclonal antibody staining was detected by using an alkaline phosphatase–antialkaline phosphatase method.^{16,17} Immunostaining was carried out with the following mAbs: anti-eosinophilic cationic protein (EG2; Pharmacia & Upjohn Diagnostics, Uppsala, Sweden), anti-CD3 (DakoCytomation Ltd, Cambridge, United Kingdom), anti-CD4 (DakoCytomation Ltd), anti-CD8 (DakoCytomation Ltd), anti-CD25 (Becton Dickinson, San Jose, Calif), anti-CD68 (clone EBM11; DakoCytomation Ltd), anti-neutrophil elastase (DakoCytomation Ltd), and anti-tryptase (clone AA1; DakoCytomation Ltd). All incubations were carried out at room temperature, and tissue sections were washed in Tris-buffered saline (TBS), pH 7.6. Monoclonal antibodies were diluted to appropriate concentrations in TBS before incubation with tissue sections (60- μL sections) for 30 minutes in a humidified slide tray, washed, and then incubated with secondary antibody (rabbit anti-mouse IgG; DakoCytomation Ltd) diluted 1:30 in 20% normal human serum (obtained from National Blood Donor Service) in TBS. Alkaline phosphatase–antialkaline phosphatase (DakoCytomation Ltd) diluted 1:30 with 20% normal human serum in TBS was incubated with the tissue sections for a further 30 minutes followed by Fast Red (Sigma Chemical Company, St Louis, Mo) diluted to 1 mg/mL in substrate buffer (0.1 M Tris, pH 8.2, containing 0.2 mg/mL naphthol ASMX phosphate and 0.1 M levamisole), filtered with a 0.2- μm syringe filter, and then incubated with the tissue sections for 20 minutes. Then sections were counterstained in Harris' Haematoxylin (Merck Ltd) for 30 seconds. Slides were washed thoroughly and mounted in Glycergel (DakoCytomation Ltd).

Basophils were detected in bronchial biopsy specimens through use of mAb 2D7 (gift from Professor L. Schwarz). Sections were immersed in 0.6% hydrogen peroxide and methanol for 30 minutes to inhibit endogenous peroxidase activity. They were then washed twice in TBS, pH 7.6, before incubation with normal horse serum (Vectastain ABC kit; Vector Laboratories, Peterborough, United

Kingdom) for 1 hour at room temperature. Sections were incubated at 4°C overnight with mAb 2D7 (0.65 $\mu\text{g/mL}$) or with the isotype-matched negative control (MOPC-31C; Sigma Chemical Company). After several washes with Tween-20 TBS (Sigma-Aldrich, Dorset, United Kingdom), sections were incubated with biotinylated horse anti-mouse IgG (Vector ABC kit; Vector Laboratories) for 1 hour at room temperature. Slides were then incubated with Vectastain avidin biotin complex for 30 minutes at room temperature. Sections were monitored microscopically during incubation with the diaminobenzidine substrate kit (Vector Laboratories). Finally, slides were washed and mounted in Glycergel (DakoCytomation Ltd).

In situ hybridization

Riboprobes, antisense, and sense were prepared from the appropriate cDNA. cDNA was inserted into different pGEM vectors and linearized with appropriate enzymes before transcription. Transcription was performed in the presence of ^{35}S -uridine triphosphate and the appropriate T7 or SP6 RNA polymerase. For *in situ* hybridization, 6- μm sections were permeabilized with Triton X-100 (Sigma-Aldrich) and then digested with proteinase K (Sigma-Aldrich). To inhibit nonspecific binding of ^{35}S probes, sections were pretreated with iodoacetamide and N-ethylmaleimide, followed by acetic anhydride-triethanolamine.¹⁸ Sections were then hybridized with either antisense probes or (control) sense probes (with an identical sequence to mRNA), as previously described.¹⁸ As a negative control, sections were treated with ribonuclease A solution before the hybridization with antisense probes.

Specific hybridization signals were recognized as clear dense deposits of silver grains in the photographic emulsion overlying cells expressing specific mRNA. The specificity of the results obtained during hybridization was confirmed by the complete absence of hybridization signals with an antisense probe (identical nucleotide sequence to the mRNA under investigation). Slides were counted blind in a random coded fashion through use of a Zeiss Axiolab microscope (Jena, Germany) fitted with an eyepiece graticule (0.202 mm^2 at a magnification of 200 $\times$).

Healthy control subjects

Six additional atopic asthmatic subjects (3 men; median age, 31.5 years; interquartile range [IQR], 24.2–43.2 years), were recruited in the study to serve as atopic control subjects. One of the 6 control subjects was sensitized to house dust mite, and the other 5 were sensitized to grass pollen.

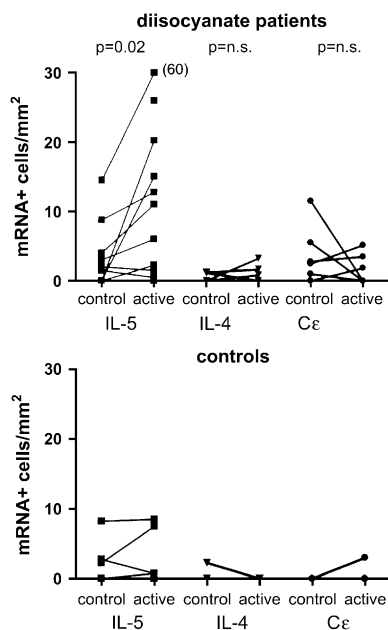


FIG 1. IL-5, IL-4, and Cε mRNA in bronchial biopsy specimens from diisocyanate patients 24 hours after an active and control challenge (upper panel) and healthy nonatopic control subjects after an active and control challenge (lower panel).

Statistical analysis of immunohistochemistry and *in situ* hybridization

The Wilcoxon matched-pairs signed-rank test was used for within-group comparisons. *P* values of less than .05 were considered statistically significant. Statistical tests were performed through use of a standard computer package (Minitab Inc, State College, Pa).

RESULTS

Occupational inhalational testing

Five of the 12 patients had a positive inhalation test response to diisocyanate, confirming occupational asthma, all with reproducible reductions in FEV₁ after active exposures. Four of the 5 (all atopic) also experienced an increase in nonspecific bronchial responsiveness. The patient with a positive inhalation challenge result but stable histamine PC₂₀ value (>16 mg/mL) after active exposure was not atopic. Four of the 7 with negative challenge results were atopic.

Cε and IL-4 message in bronchial biopsy specimens

There was a striking deficiency in Cε and IL-4 mRNA-positive cells in bronchial biopsy specimens from patients challenged with diisocyanate (Cε median of 0 and IQR of 0-1.85; IL-4 median of 0 and IQR of 0-0.85); this was the case both for patients with positive and negative inhalation challenge test results (Fig 1 and Table III). Resting levels of Cε mRNA-positive cells were greater in some diisocyanate-exposed patients (median of 0 and IQR of 0-2.75) when compared with those of healthy control subjects (median

of 0 and IQR of 0-0), although these were predominantly in those with negative inhalation test responses. There was no difference in the number of Cε and IL-4 mRNA-positive cells in either the positive or negative challenge groups.

In contrast to patients exposed to diisocyanate, atopic asthmatic subjects who underwent segmental bronchial allergen challenge with house dust mite or grass pollen showed significant increases in Cε RNA (median of 0/mm² [IQR of 0-10.35] before and median of 25.9/mm² [IQR of 19.9-61.6] after allergen challenge; *P* = .036) and IL-4 mRNA-expressing cells (median of 0/mm² [IQR of 0-0.5] before and median of 10.8/mm² [IQR of 4.8-37.2] after allergen challenge; *P* = .059) in the bronchial mucosa after allergen challenge compared with the control challenge.

IL-5, CD25, CD4, and eosinophils in bronchial biopsy specimens

Increased numbers of IL-5-, CD25- (IL-2 receptor-bearing cells), and CD4-positive cells and a trend toward an increase in eosinophils were found after active challenge in the bronchial mucosa of patients exposed to diisocyanate but not in healthy control subjects (Fig 2). The number of cells expressing IL-5 mRNA increased significantly between the active and control challenge in the diisocyanate patients (*P* = .02, Table II). The number of CD4- and CD25-positive cells and eosinophils also increased in diisocyanate patients after an active challenge, although these changes were not statistically significant (Table II). There was evidence of an increase in the number of elastase-positive cells in the bronchial mucosa of diisocyanate patients after active challenge (Table II).

We were unable to detect increases in IL-13 and IFN-γ mRNA after active challenge in diisocyanate patients (Table II). Similarly, the numbers of CD68- or AA1-positive cells were not increased after active challenge (Table II). There were no significant differences in any inflammatory markers between patients with a positive or negative diisocyanate challenge result, either after a control or an active challenge.

DISCUSSION

To our knowledge, this is the first study to investigate the presence of Cε message in the bronchial mucosa of patients with diisocyanate-induced asthma. We found a striking absence of both bronchial Cε and IL-4 message after inhalation challenge with diisocyanate, irrespective of whether the challenge test result was positive or negative. We propose that this finding is suggestive that IgE is not necessarily associated with the induction of occupational asthma by diisocyanates, at least in patients in whom serum-specific IgE antibodies to diisocyanate are undetectable.

The lack of Cε message in the bronchial mucosa of patients with diisocyanate-induced asthma is consistent with the absence of a detectable concentration of diisocyanate-specific IgE antibodies in their serum. The lack of

TABLE III. Median expression and phenotype of cells after control and active diisocyanate challenges in patients

	Control challenge, median (IQR)	Active challenge, median (IQR)	P value
<i>In situ</i> hybridization			
Cε	0 (0-2.7)	0 (0-1.8)	.68
IL-5	2.0 (0-4.0)	11.0 (1.5-20.2)	.02
IL-4	0 (0-1.1)	0 (0-0.8)	1.0
IL-13	3.5 (1-5.5)	2.5 (0-4.2)	.55
IFN-γ	0 (0-2)	1.5 (0-5.5)	.9
Immunohistology			
EG2	5.2 (1.3-18.7)	25.5 (3.6-56.8)	.10
CD25	4.0 (3.5-8.3)	22.0 (1.2-43.3)	.34
CD4	90 (33.0-139.0)	103 (38.0-200.0)	.07
CD68	112.5 (75.0-125.0)	121.5 (49.0-223.5)	.54
CD3	236.0 (199.0-329.0)	238.0 (135.0-407.0)	.92
Elastase-positive cells	24.5 (16.3-68.2)	64.0 (39.4-76.6)	.07
CD8	164 (40.8-179.5)	129 (80.0-176)	.22
AA1	33.3 (22.4-53.5)	27.3 (22.3-45.1)	.84

expression of Cε RNA is, however, in contrast to the findings from patients with both atopic and nonatopic asthma unrelated to work.¹² Similarly, the lack of IL-4 is at variance with the responses of patients challenged with high-molecular-weight allergens¹⁹ and those with either atopic or nonatopic asthma.^{12,20} One report identified IL-4-producing cells in the bronchial mucosa of patients with diisocyanate-induced asthma 48 hours after active challenge but not at 1 to 4 weeks after the last workplace exposure.²¹ We doubt we have missed the presence of IL-4 (measured 24 hours after challenge) because we detected IL-4 mRNA 6 hours after challenge in nasal biopsy specimens from patients with allergic rhinitis.²² In T-cell clones derived from the bronchial mucosa of patients with diisocyanate-induced occupational asthma, IL-4 was detected in only 6% of clones.²³ Specific IgE antibodies to diisocyanates were not determined in these patients.

In our study the numbers of both CD4 and CD25 cells were increased, suggesting that activated T cells play a role in the development of diisocyanate-induced asthma as they do in both extrinsic and intrinsic disease.²⁴ The number of cells expressing IL-5 message was significantly increased after diisocyanate challenge. These findings are complementary to those of Maestrelli et al,²¹ who observed an increase in IL-5 at the protein level in patients with diisocyanate-induced asthma after challenge. IL-5 promotes differentiation, vascular endothelial adhesion, and survival of eosinophils,²⁵ and although the changes were not statistically significant, the numbers of eosinophils were increased in the bronchial mucosa after active challenge in our diisocyanate-exposed patients. These results are comparable with those of other studies of diisocyanate-induced asthma²⁶ and mild atopic asthma.¹⁹ Eosinophil recruitment and activation are likely to be important in asthma of diverse causes.

This study has highlighted important similarities and differences between diisocyanate-induced occupational asthma and both atopic and intrinsic asthma. In common with atopic and intrinsic asthma, patients with

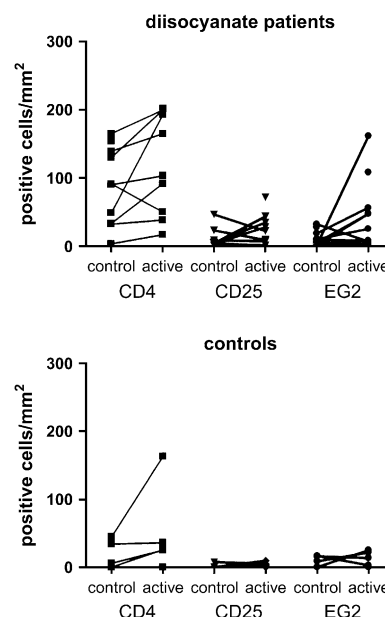


FIG 2. CD4-, CD25-, and EG2-positive cells in bronchial biopsy specimens from diisocyanate patients 24 hours after an active and control challenge (*upper panel*) and healthy nonatopic control subjects after an active and control challenge (*lower panel*).

diisocyanate-induced asthma have a significant increase in IL-5 after an active challenge and increased numbers of activated T cells. In contrast, patients with diisocyanate-induced asthma, unlike patients with both atopic and intrinsic asthma, have no detectable Cε or IL-4 after active challenge with allergen. This suggests that patients with diisocyanate-induced asthma might have different mechanisms from both atopic and intrinsic asthma.

Previous reports have proposed that diisocyanate-induced asthma might be due to a non-IgE-mediated mechanism in some cases.¹¹ Neuroimmune interactions in the lung have been reported in animal models of TDI-

induced asthma.²⁷⁻²⁹ Recently, TDI was found to cause an influx of inflammatory cells, particularly neutrophils, and an increased production of IL-8, followed by an increase in substance P in sensory neurons.³⁰ Diisocyanates are believed to bind to airway epithelial cell proteins and are taken up by these cells.^{31,32} Exposure to diisocyanates leads to the local formation of reactive oxygen species that are used by enzymes of the glutathione S-transferase supergene family. Intracellular S-glutathionyl adducts have been identified in murine lung *in vivo* and in human bronchoepithelial cells *in vitro* after exposure to TDI,³¹ suggesting that these enzymes have a biologic role in TDI-induced inflammatory pathology. Further evidence comes from genetic studies of glutathione S-transferase polymorphisms. Lack of the GSTM1 gene was associated with almost a 2-fold risk of diisocyanate-induced asthma. The combination of GSTM1 null and GTM3 AA phenotype was strongly associated with lack of diisocyanate-specific IgE antibodies and with late-phase responses after bronchial provocation.³³

In our patients with diisocyanate-induced asthma without detectable specific IgE antibodies in their serum, there was no evidence of local bronchial IgE production after isocyanate exposure sufficient to provoke an asthmatic reaction. This suggests that occupational asthma caused by diisocyanates is an IgE-independent disease, at least in a proportion of patients. Similarly, the failure to detect specific IgE antibodies in serum might not be due to a technical issue of inappropriate conjugation.³⁴ We hypothesize that there are 2 separate forms of asthma induced by diisocyanate exposure, one associated with and in part mediated through IgE production and the other not. There is some indirect evidence to support this hypothesis. Patients with diisocyanate-induced asthma and detectable IgE have fewer symptoms of asthma, use less medication, and have a significantly shorter duration of symptoms, latency, and duration of exposure to diisocyanates compared with those patients without specific IgE antibodies.³⁵ These differences in the induction and progression of asthma might result from particular genotypes: the combination of glutathione S-transferase genes GSTM1 and GSTM3 is associated with a lack of diisocyanate-specific IgE antibodies.³³ Furthermore polymorphisms in the glutathione S-transferase gene GSTP1 is associated with susceptibility to diisocyanate-induced asthma induced by prolonged exposure to TDI, suggesting that these genes might modulate the development of the underlying pathology.³⁶ To test our hypothesis further, the findings from this study need to be extended to a further group of patients with diisocyanate-induced asthma with detectable specific IgE antibodies.

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