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Airway inflammation in occupational asthma caused by styrene

To the Editor:

Styrene or phenylethylene has rarely been reported as a causative agent of occupational asthma.¹⁻⁴ This volatile monomer is widely used in the production of polymers, copolymers, and reinforced plastics and is a reactive solvent in the manufacture of unsaturated polyester resins. Styrene is known as an irritant to the skin and mucous

membranes and has been associated with dysfunction of the central nervous system. Workers in auto body shops may be exposed to styrene, because they use polyester resins in paints and car putty.

A 31-year-old man, a nonatopic and nonsmoker with persistent asthma, was referred for evaluation to the outpatient allergy clinic of our hospital. He had worked as a painter in an auto body shop for 4 years on a regular 8-hour shift during the day. He did not wear any protective equipment. During the last year, he had experienced sneezing, nasal discharge, ocular itching, and nasal stuffiness as well as dyspnea, dry cough, and wheezing when working. Asthma symptoms were more intense at the end of the work shift and at night, and he remained symptom-free during holidays and days off work. He handled paints containing hexamethylene diisocyanate (HDI) and polyester resins and hardeners that included in their composition styrene at a concentration of 10% to 25%, as disclosed by product labels and material data safety sheets.

He was evaluated 4 months after he had left his job because of asthma symptoms. After obtaining written informed consent, 3 single-blind specific inhalation challenges (SICs) were performed in a 7-m³ challenge chamber on separate days. This chamber is made of lacquered stainless steel (Telstar Projects SA, Madrid, Spain) and equipped with vacuum exhaust and high-efficiency particle arrest filtration. On visit 1, PC₂₀ was measured, and sputum induction was performed. On visit 2, the patient painted placebo solution (polyol) with a brush onto a cardboard for 5 minutes. Spirometry was performed at 5, 10, 20, 30, 40, and 60 minutes after the inhalation challenge. From that moment, peak expiratory flow and FEV₁ were measured hourly with a computerized flowmeter (Amos, Jaeger, Germany) for 24 hours after challenge, respecting sleeping time. On visit 3, the patient underwent a SIC with HDI. Twenty-four hours later, on visit 4, methacholine inhalation test and sputum induction were performed again. Three days later, on visit 5, the patient painted the styrene solution (20 mL) on a cardboard for 4 minutes, and the following day, on visit 6, a methacholine inhalation test and sputum induction were performed.

Physical examination, blood tests, and spirometry results were normal. Specific IgE against diisocyanates measured in the patient's serum was negative. Specific IgE against styrene was assayed in duplicate by ELISA, using a concentration of 20 µg/mL and serum dilution series starting from 1:2 as described elsewhere,⁵ and was also negative. Sputum was induced by hypertonic saline inhalation and processed as previously described (both with and without dithiothreitol).⁶ Sputum samples were analyzed using flow cytometer scan cytometry (FACSCalibur flow cytometer; Becton Dickinson, San José, Calif). mAbs (Pharmingen, San Diego, Calif) used to identify cells were CD19, CD3, CD4, CD8, CD16, CD45, CCR3 (B and T lymphocytes, neutrophils, and eosinophils), CD63, and CDw125 (activated basophils).

At baseline (visit 1), PC₂₀ methacholine was 1 mg/mL. On visit 2 (sham exposure), no significant changes in FEV₁ were observed over the next 24 hours. On visit 3,

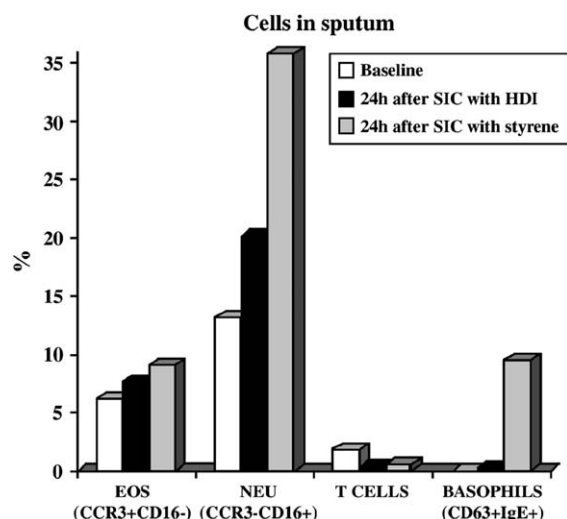


FIG 1. Comparison of differential cell counts in sputum at baseline and after specific inhalation challenge with HDI and styrene. Sputum eosinophils (CCR3⁺CD16⁻) and activated basophils (CD63⁺IgE⁺) increased 24 hours after styrene challenge compared with baseline and HDI challenge. EOS, Eosinophils; NEU, neutrophils.

SIC with HDI at a concentration of 15 ppb for 120 minutes elicited no reaction. No significant changes in FEV₁, PC₂₀ methacholine, or cell counts in induced sputum were observed 24 hours after the HDI challenge. On visit 5, the styrene challenge elicited an isolated late asthmatic response, with a maximum fall in FEV₁ of 22% 11 hours after the provocation test. An increase in sputum eosinophils from 0.02×10^6 to 0.43×10^6 cells/mL and a striking increment of activated basophil counts in induced sputum were observed 24 hours after styrene SIC (Fig 1), but no significant changes were observed in PC₂₀ methacholine. The results of induced sputum analysis by flow cytometry did not vary with or without dithiothreitol.

Three subjects with suspected occupational asthma underwent the same challenge protocol with styrene, and no relevant changes in FEV₁ or PC₂₀ methacholine were observed (Table I). The increase in eosinophil and basophil counts in sputum after styrene SIC in the patient, as well as the negative results obtained with exposure to placebo and HDI, support the specificity of the airway reaction to styrene. A rise in eosinophil counts in sputum greater than 0.26×10^6 cells/mL after SIC⁷ or greater than 2% after exposure at work⁸ are considered to be clinically significant. An increase in basophil counts in induced sputum has been found in atopic subjects with asthma⁹ and subjects with occupational asthma¹⁰ after SIC. Interestingly, the number of sputum basophils correlated with airway hyperresponsiveness to methacholine.⁹

Moscato et al² reported 2 workers with work-related asthma symptoms on exposure to styrene, and in both cases, SIC with styrene elicited early asthmatic reactions. Hayes et al³ reported an air frame technician with occupational asthma caused by styrene in whom SIC with this agent provoked a dual asthmatic response and increased

TABLE I. PC₂₀ methacholine before and after SIC in the patient and 3 control subjects

	Baseline PC ₂₀ (mg/mL)	PC ₂₀ 24 h after SIC (mg/mL)	Maximum drop in FEV ₁ (%)
Patient			
HDI SIC	1.0	2.3	5
Styrene SIC	2.3	2.7	22
Control 1			
Styrene SIC	1.8	2.7	3
Control 2			
Styrene SIC	1.7	2.0	5
Control 3			
Styrene SIC	>16	>16	4

responsiveness to inhaled histamine. However, evidence of lower airway inflammation had not been previously documented with styrene exposure. Öner et al⁴ surveyed 47 workers of the furniture industry with a history of exposure to styrene. Although 3 workers had a positive methacholine test and 1 of them had a peak expiratory flow record compatible with occupational asthma, SIC with styrene in these 3 workers was negative.

Styrene is increasingly being used for the manufacture of a broad range of materials. This agent may elicit upper and lower airway symptoms as well as airway inflammation. Although the underlying mechanism causing these events is still unknown, it appears to be similar to other low-molecular-weight agents causing immunologic occupational asthma. The causative role of styrene in occupational rhinitis and asthma should be investigated among workers exposed to this compound.

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Prolonged elevation of serum tryptase in idiopathic anaphylaxis

To the Editor:

Idiopathic anaphylaxis is a well described type of anaphylaxis for which no identifiable provocative agent exists.¹ Serum tryptase levels, a marker of mast cell activation, can be used in the diagnosis of anaphylaxis.² Only 1 known case report exists of elevated tryptase levels in a patient who had a single episode of idiopathic anaphylaxis.³ We describe the case of a patient with idiopathic anaphylaxis and an elevated mature tryptase level ~26 hours after the onset of symptoms.

The patient is a 54-year-old man who presented for office evaluation after having episodes of anaphylaxis for more than 10 years. Symptoms during his episodes included urticaria, pruritus, facial and extremity angioedema, nausea, vomiting, diarrhea, lightheadedness, syncope, and documented hypotension. The severity of his episodes was variable from mild symptoms of angioedema to more severe anaphylaxis with urticaria, angioedema, and hypotension necessitating emergency department admissions. The frequency of his episodes varied between 4 and 18 per year, with an apparent increase in frequency the year before his evaluation. History failed to reveal an inciting factor, including exercise, foods, medications, and insect stings. His past medical history was notable only for hypertension treated with amlodipine. His physical examination while asymptomatic was remarkable only for minimal nasal turbinate edema. Skin prick tests to a panel of 79 foods and spices were negative. Food dye and preservative challenges were also negative. Complete blood count with differential, general chemistries, C1 esterase inhibitor, and complement levels were normal.

Based on his history and laboratory evaluation, a diagnosis of idiopathic anaphylaxis was made. The patient was initially instructed to have a serum tryptase level measured with his next episode. Two months later, approximately 3 hours after eating breakfast, he developed pruritus and urticaria, mild swelling of his hands and feet, and

periorbital and lip edema, followed by throat tightness and severe gastrointestinal distress including nausea, vomiting, and diarrhea. He also experienced lightheadedness that was orthostatic in nature. He received no medical treatment for this episode and simply stayed at home. In the past, the patient had used self-injectable epinephrine, but he found the relief very short-lived and therefore stopped using it.

During this episode, he began a 24-hour urine collection for methylimidazoleacetic acid. His methylimidazoleacetic acid was elevated at 10.1 mg/24 h (normal < 5). Previous baseline 24-hour urine collection for histamine was normal.

The next day, his symptoms had resolved with the exception of a few urticarial lesions. Approximately 26 hours after the initial episode began, a serum tryptase level was measured at 110 ng/mL mature (β) tryptase (normal mature tryptase < 1 ng/mL). All symptoms with the exception of urticaria had resolved by the time this serum sample was obtained. Baseline serum obtained 6 months and 2 years later revealed mature tryptase levels of < 1 ng/mL and total tryptase levels of 2 and 3 ng/mL, respectively (normal total tryptase, 1-15 ng/mL). All tryptase assays were performed at Virginia Commonwealth University.

To our knowledge, this is just the second case report of an elevated serum tryptase level in a patient with idiopathic anaphylaxis. In the previous case report, no mention of baseline tryptase levels or follow-up was made.³ The fact that our patient had transient elevations of mature (β) tryptase and a urinary histamine metabolite with an acute episode and normal levels of both mature and total tryptase while asymptomatic strengthens the diagnosis of anaphylaxis involving mast cell activation and makes systemic mastocytosis unlikely. This patient has now been followed for 10 years, and although the frequency of his anaphylactic episodes has decreased substantially, no specific etiology has been determined. The mechanism of idiopathic anaphylaxis has yet to be elucidated, but it is important to make the diagnosis accurately early in the disease course, because the condition is known to recur.¹ Although further investigation is needed, serum tryptase levels obtained during or even after an episode of idiopathic anaphylaxis may aid in making the diagnosis correctly.

Previously, it has been shown that the half-life of serum mature tryptase is about 2 hours, and it has been suggested that tryptase should be checked within 4 hours of an anaphylactic episode.⁴ This case is unique because the elevated tryptase level was drawn ~26 hours after the onset of symptoms and approximately 12 hours after resolution of all symptoms with the exception of a few urticarial lesions. This suggests that in some individuals with anaphylaxis, a serum tryptase may be elevated well beyond this 4-hour interval. In some patients suspected of having anaphylaxis, serum samples may be available only more than 4 hours after the episode. In such cases, determination of periacute and baseline tryptase levels may indeed be useful in securing a diagnosis of anaphylaxis involving mast cells.⁵