

Beutler and Beutler further state that correlations may merely reflect a linkage with nearby mutations. This possibility cannot be ruled out, as we discussed in our article, but it seems unlikely, because the functional significance of the TLR4 polymorphism is well documented in vivo and in cellular transfection studies,¹ and the experimental and epidemiologic findings are highly consistent.

Finally, Beutler and Beutler present data from a small case-control study comparing patients who underwent carotid endarterectomy with controls. Even if we assume that this study is free from bias, that it is adequately powered, and that our Asp299Gly and their TLR4 promoter polymorphisms are functionally equivalent, the findings are not in conflict with ours, because they tested the wrong hypothesis: pathogenic mechanisms underlying vessel stenosis are substantially different from those involved in atherogenesis. We have previously shown that the development of carotid stenosis relies primarily on atherothrombosis and on procoagulant risk factors^{2,3} and that this advanced stage in vessel disease is not significantly related to inflammation and infectious illness.⁴

STEFAN KIECHL, M.D.
JOHANN WILLEIT, M.D.
University of Innsbruck
A-6020 Innsbruck, Austria

DAVID A. SCHWARTZ, M.D.
Duke University Medical Center
Durham, NC 17710

Editor's note: Dr. Schwartz reports receiving research support from Eisai Research Institute.

1. Arbour NC, Lorenz E, Schutte BC, et al. TLR4 mutations are associated with endotoxin hyporesponsiveness in humans. *Nat Genet* 2000;25:187-91.
2. Kiechl S, Willeit J. The natural course of atherosclerosis. I. Incidence and progression. *Arterioscler Thromb Vasc Biol* 1999;19:1484-90.
3. Willeit J, Kiechl S, Oberhollenzer F, et al. Distinct risk profiles of early and advanced atherosclerosis: prospective results from the Bruneck Study. *Arterioscler Thromb Vasc Biol* 2000;20:529-37.
4. Kiechl S, Egger G, Mayr M, et al. Chronic infections and the risk of carotid atherosclerosis: prospective results from a large population study. *Circulation* 2001;103:1064-70.

Bronchiolitis in Popcorn-Factory Workers

To the Editor: Kreiss et al. (Aug. 1 issue)¹ report a high incidence of bronchiolitis obliterans at a microwave-popcorn factory. The chemical diacetyl (2,3-butanedione) was singled out as a possible causal agent of this deadly condition and other medical problems found in workers in this plant. As a chemist, biochemist, and toxicologist, I would like to point out that 2,3-butanedione is in chemical equilibrium with 1,3-butane-diene-2,3-diol (Fig. 1, facing page). This phenomenon, which is well known in organic chemistry, is called keto-enol tautomerism. This isomer is expected to be very reactive with oxygen both at room temperature and on heating. Thus, 1,3-butane-diepoxyde-2,3-diol would be expected as a product. Although the parent compound is known to be reactive with arginine, the diepoxyde

is of particular interest, since butadiene diepoxyde is a known human carcinogen. The appropriate government agencies must investigate and evaluate whether diacetyl should be banned from food products.

EDWARD G. EZRAILSON, PH.D.
2308 West Settler's Way
The Woodlands, TX 77380
edez1@prodigy.net

1. Kreiss K, Gomas A, Kullman G, Fedan K, Simoes EJ, Enright PL. Clinical bronchiolitis obliterans in workers at a microwave-popcorn plant. *N Engl J Med* 2002;347:330-8.

To the Editor: Kreiss and colleagues report frequent cases of bronchiolitis obliterans among workers in a popcorn plant that were attributed to the inhalation of the volatile ingredient diacetyl in the butter flavoring. Although this conclusion is in keeping with the toxic effects of diacetyl on the respiratory epithelium in animals, and although a dose-response relation (a decreasing forced expiratory volume in one second associated with increasing exposure to diacetyl) was established, it may not be the only causative agent. The workers who were affected the most were also exposed to the highest concentrations of other volatile compounds and respirable dust. Maize bran, glumes, and stigmas contain considerable amounts of tannins,¹ which are necessarily constituents of airborne particles. Inhaled tannins are considered to be an important causal factor in obstructive pulmonary diseases among workers exposed to dust of plant origin, such as those who work in cotton mills or grain elevators and those who work with herbal tea.² Therefore, tannins may be one of the substances implicated in the development of "popcorn worker's lung." This hypothesis is supported by the finding that clinical symptoms that follow the occupational inhalation of tannins are similar to those reported among popcorn workers. In addition, the lack of improvement in the symptoms with β_2 -agonist bronchodilators is consistent with the inhibitory effect of tannin on adenylate cyclase in airway epithelial cells.³

DIRK TAUBERT, M.D., PH.D.
ANDREAS LAZAR, M.D.
EDGAR SCHÖMIG, M.D.
Medical College of the University of Cologne
50931 Cologne, Germany
dirk.taubert@medizin.uni-koeln.de

1. Bradley PR, ed. *British herbal compendium*. Vol. 1. Bournemouth, England: British Herbal Medicine Association, 1992.
2. McL Niven R, Pickering CA. Byssinosis: a review. *Thorax* 1996;51:632-7.
3. Cloutier MM, Guernsey L. Tannin inhibits adenylate cyclase in airway epithelial cells. *Am J Physiol* 1995;268:L851-L855.

To the Editor: In his editorial (Aug. 1 issue),¹ Schachter comments on occupational airway diseases but leaves out what I think is an important finding described in the accompanying article by Kreiss et al. As the occupational physician involved in this case, I noted that not only was an epidemic of bronchiolitis obliterans present, but the number of tobacco smokers involved was unusually small.²

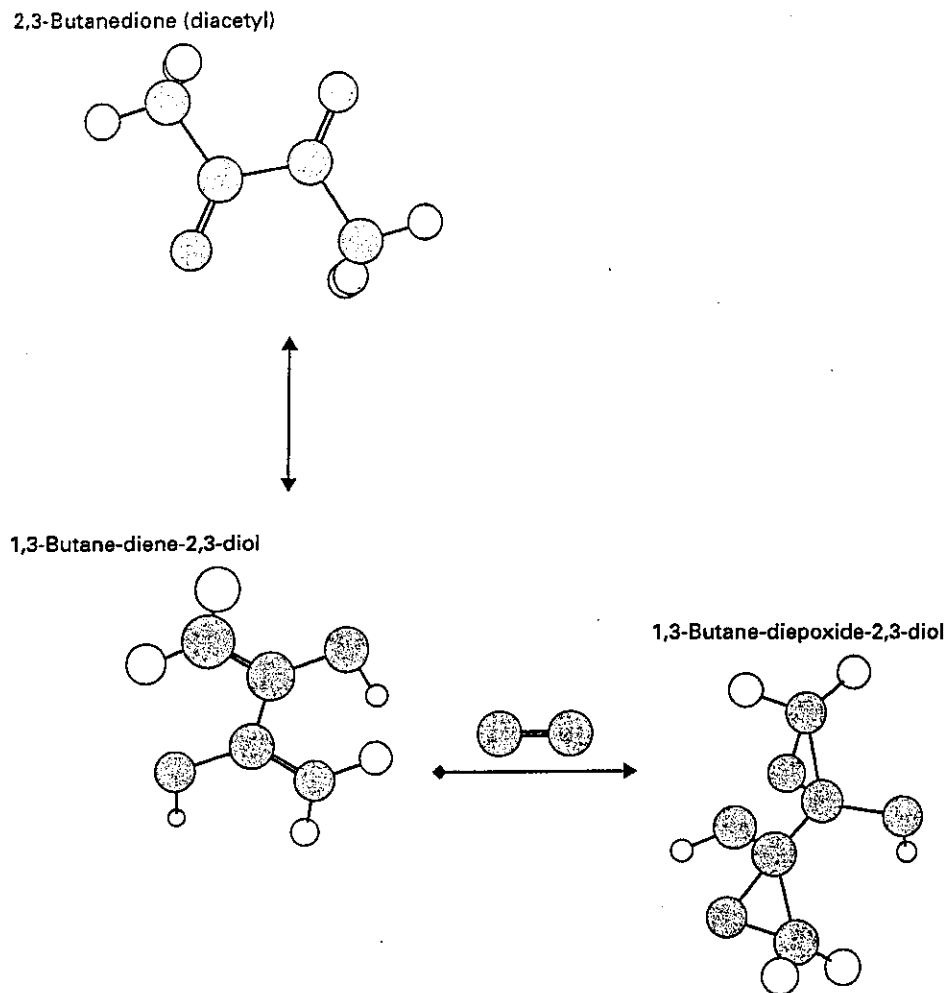


Figure 1. Chemicals 2,3-Butanedione and 1,3-Butane-Diene-2,3-Diol, and Their Expected Product, 1,3-Butane-Diepoxyde-2,3-Diol.

Only one of the initial eight patients was a smoker. Non-smokers were overrepresented among patients as compared with the exposed population. In the study population described by Kreiss et al., the workers who never smoked had a rate of airway obstruction that was three times as high as that among the smokers, although all workers were affected. An understanding of the mechanism of this protection could lead to preventive interventions.

ALLEN J. PARMET, M.D., M.P.H.
Midwest Occupational Medicine
Kansas City, MO 64108
mommd@kc.rr.com

- Schachter EN. Popcorn worker's lung. *N Engl J Med* 2002;347:360-1.
- Parmer AJ, Von Essen S. Rapidly progressive, fixed airway obstructive disease in popcorn workers: a new occupational pulmonary illness? *J Occup Environ Med* 2002;44:216-8.

The authors and a colleague reply:

To the Editor: We used diacetyl as an index of exposure to volatile organic chemicals in the popcorn plant because it was the predominant one found in plant air. However, identification of the causal agent or agents in the flavoring will rely on studies in animals in which individual constituents are tested; such studies are now under way. Diacetyl is a leading candidate for investigation of potential respiratory toxicity because alpha-dicarbonyl compounds react with functionally reactive arginine residues in proteins and with guanine and inhibit superoxide dismutase and glutathione reductase, which are involved in protection from oxidative stress. In addition to Dr. Ezrailson's concern about the properties of a derivative diepoxyde, diacetyl itself has been nominated for studies by the National Toxicology Program (NTP) because of widespread human exposure, limited evidence of mutagenicity, and relations to

carcinogens and mutagens in terms of structure and activity, as well as because diacetyl is representative of aliphatic alpha-diketones. (See the NTP Web site at <http://ntp-server.niehs.nih.gov>.)

We did not detect 1,3-butadiene-2,3-diol or 1,2;3,4-diepoxybutane-2,3-diol in any samples collected by thermal desorption tubes and analyzed with gas chromatography-mass spectrometry. However, we agree with Dr. Ezrailson that diacetyl would be present in equilibrium with its tautomers, as governed by the equilibrium constants for the conversions. Since diacetyl occurs naturally in butter and during the manufacture of alcoholic beverages, any proposed ban of diacetyl in food products raises issues of practicality.

As noted by Taubert and colleagues, other agents within the workplace may contribute to the clinical bronchiolitis obliterans seen in this workforce. Indeed, necrosis of the respiratory epithelium in the mainstem bronchus was more severe in rats exposed to butter-flavoring vapors than in rats exposed to diacetyl alone at a similar diacetyl concentration (unpublished data). We did not measure tannins. Workers managing the grain bins, presumably with greater exposure to organic dust, were in the low-risk group; mixers, who had almost no active contact with corn or its dusts, had the highest historical risk of fixed airway obstruction. The role of respirable salt dust in the airway damage found in microwave-popcorn production workers remains unclear. However, our observation that the same syndrome occurs in flavoring-production workers without exposure to grains or salt makes these agents less likely to be causal contributors.

KATHLEEN KREISS, M.D.
ANN HUBBS, D.V.M., PH.D.
GREGORY KULLMAN, PH.D.

National Institute for Occupational Safety and Health
Morgantown, WV 26505
kkreiss@cdc.gov

The editorialist replies:

To the Editor: Dr. Parmet points out an interesting but unexplained observation of his study and that by Kreiss et al. In his original study, nonsmoking workers accounted for the majority of index cases of bronchiolitis; among the workers studied by Kreiss et al., those who had never smoked had unusually high rates of airway obstruction. This latter finding is not particularly unusual, since a high prevalence of disease among nonsmokers is frequently used to confirm the presence of a true occupational or environmental effect.¹ What Parmet focuses on is the fact that although the frequency of airway obstruction in smoking workers in this cohort is increased (prevalence ratio, 1.6), it is not increased to the same extent as that among nonsmoking workers (prevalence ratio, 10.8). In occupational airway disease, the effect of the pollutant tends to be more pronounced among smokers, because the injury is frequently additive. Possible explanations for the lack of such an additive effect in this setting include a healthy-worker effect, by which sicker smoking workers would leave the industry at an early date, before the onset of bronchiolitis, and the possibility that cigarette smoking alters the

deposition of inhaled particles² in such a way as to decrease the amount of other pollutants arriving in smaller airways. Further speculation is possible, but the primary public health message raised by these studies remains clear: injury to the airway in industries dealing with organic pollutants such as those associated with the manufacturing of microwave popcorn may be frequent, disabling, and occasionally life-threatening.

E. NEIL SCHACHTER, M.D.
Mount Sinai School of Medicine
New York, NY 10029

1. Beck GJ, Maunder LR, Schachter EN. Cotton dust and smoking effects on lung function in cotton textile workers. *Am J Epidemiol* 1984;119:33-43.
2. Lippmann M, Yeates DB, Albert RE. Deposition, retention, and clearance of inhaled particles. *Br J Ind Med* 1980;37:337-62.

Inflammatory Bowel Disease

To the Editor: In his review of inflammatory bowel disease (Aug. 8 issue),¹ Podolsky refers to interleukin-10 as a down-regulatory cytokine, citing findings in murine models. However, recent data do not support an antiinflammatory role for interleukin-10 in patients with inflammatory bowel disease. In this regard, Tilg et al. reported that recombinant interleukin-10 administered to patients with Crohn's disease increased the production of interferon- γ .² The inflammatory role of interleukin-10 found in their study paralleled the absence of significant benefits in clinical trials assessing the efficacy of high doses of recombinant human interleukin-10.^{3,4} Such data reveal the complexity of the cytokine network in human inflammatory bowel disease, as well as the need for applying the findings of basic research to clinical practice cautiously.

JAIME GARCÍA DE TENA, M.D., PH.D.
Hospital Universitario Príncipe de Asturias
28805 Alcalá de Henares, Spain
jgtena@terra.es

LUIS MANZANO ESPINOSA, M.D., PH.D.
Hospital Universitario Ramón Cajal
28034 Madrid, Spain

MELCHOR ALVAREZ-MON, M.D., PH.D.
Hospital Universitario Príncipe de Asturias
28805 Alcalá de Henares, Spain

1. Podolsky DK. Inflammatory bowel disease. *N Engl J Med* 2002;347:417-29.
2. Tilg H, van Montfrans C, van den Ende A, et al. Treatment of Crohn's disease with recombinant human interleukin 10 induces the proinflammatory cytokine interferon gamma. *Gut* 2002;50:191-5.
3. Schreiber S, Fedorak RN, Nielsen OH, et al. Safety and efficacy of recombinant human interleukin 10 in chronic active Crohn's disease. *Gastroenterology* 2000;119:1461-72.
4. Fedorak RN, Gangl A, Elson CO, et al. Recombinant human interleukin 10 in the treatment of patients with mild to moderately active Crohn's disease. *Gastroenterology* 2000;119:1473-82.