Assessment of risk factors for IgE-mediated sensitization to tetrachlorophthalic anhydride

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Two groups of workers with occupational asthma caused by tetrachlorophthalic anhydride (TCPA) have been reported on; in only one report was specific anti-TCPA antibody demonstrated. We received a request to investigate respiratory problems among workers at a plant where solenoid coils are manufactured with the use of epoxy resin and where a new TCPA hardener had been introduced in 1988. In response, we conducted a cross-sectional evaluation of 52 current workers and assayed serum samples from 49 workers for TCPA–human serum albumin–specific IgE and IgG by ELISA. Mean airborne TCPA concentrations ranged from 0.21 to 0.39 mg/m³ in 1989 and May 1990 but were considerably lower (less than 0.009 to 0.13 mg/m³) by July 1990 when ventilation was introduced. Workers were classified in four departments as molders (most directly working with TCPA), those intermittently exposed to TCPA, coil assemblers, and office workers. There was a high prevalence of reported work-related respiratory symptoms (from 27% to 39%). The mean decline in FEV_1 over the shift in the molders (down 4.1%) was significantly different from the change in the office group (increased 2.5%). Intrashift change in FEV_1 was inversely correlated with log personal TCPA concentration ($r = -0.47$). Of 49 current workers, 15 (31%) had elevated serum TCPA–human serum albumin–specific IgE antibody levels and 19 (39%) had elevated specific IgG levels. The prevalence of specific IgE was highest in the mold and intermittent groups (54%), in comparison with the coil assembly (25%) and office (0%) groups. Those workers who were IgE positive were significantly less likely to be current smokers and had worked significantly longer since 1988. Of those hired in 1988 or after, the proportion of workers who were IgE positive increased with duration of employment category (p value for trend 0.022). Since ventilation was installed and TCPA exposures reduced to less than 0.1 mg/m³, there has been a marked decrease in symptoms and no new cases of occupational asthma among newly hired workers at the plant. (J ALLERGY CLIN IMMUNOL 1993;92:237-47.)

Key words: *Acid anhydrides, tetrachlorophthalic anhydride, occupational asthma, sensitization, cross-sectional studies, smoking*

Acid anhydrides are a class of chemical agents recognized to cause occupational asthma (OA).[1] Two clusters of cases of OA caused by tetrachlorophthalic anhydride (TCPA) have been reported; one in the United States,[2] the other in the United Kingdom.[3-6] Schlueter et al.[2] were unable to demonstrate specific precipitating antibody or specific IgE to a TCPA-protein antigen. Howe et al.[3] later demonstrated immediate skin prick test reactivity and serum specific IgE to a TCPA–human serum albumin (HSA) conjugate. High prevalences of serum specific IgE responses to

Abbreviations used

CI:	Confidence interval
DL:	Detection limit
FVC:	Forced vital capacity
HSA:	Human serum albumin
LRT:	Likelihood ratio test
OA:	Occupational asthma
OD:	Optical density
TCPA:	Tetrachlorophthalic anhydride
TDI:	Toluene diisocyanate
WRA:	Work-related asthma

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Received for publication Sept. 3, 1992; revised Dec. 16, 1992; accepted for publication Feb. 5, 1993.

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0091-6749/93 $1.00 + .10 **1/1/46302**

chemical-protein antigens have been reported among workers exposed to phthalic and hexahydrophthalic acid anhydrides.

In April 1990 the Ontario Ministry of Labour received a request from plant management for assistance regarding breathing problems among workers at a plant in Ontario that manufactures solenoid coils. Respiratory problems were first recognized in 1988; four index cases were evaluated by a pulmonary consultant. The objectives of the investigation were to document the extent of TCPA exposure, determine the prevalence of work-related respiratory symptoms, and determine possible risk factors associated with IgE-mediated sensitization to TCPA.

PROCESS

The facility, which employed approximately 50 workers during 1990, had manufactured solenoid coils since 1984. These are manufactured by winding copper wire on bobbins, soldering the copper wires to terminals, mechanically crimping lead wires, placing coils into metal fixtures to form the solenoid, and then embedding the metal fixture into an epoxy resin casing. The copper wires are soldered by dipping them into unheated rosin flux (49% isopropyl alcohol, 50% colophony) and then into a small lead solder pot (60% tin, 40% lead) heated to 950° F (510° C).

The epoxy resin, which contains TCPA and other constituents including silica, is received in bags of granules that are compressed into pills by a preformer machine. The pills are then preheated and placed inside a molding machine, together with the solenoid assembly. Molding then takes place for 3.5 minutes at 330° to 350° F (166° to 177° C). After molding is completed the parts are deflashed, inspected, and marked. Marking is done either by hot stamping the epoxy casing or by printing with a two-component ink containing 0.7% toluene diisocyanate (TDI). Early in 1988, a new epoxy resin hardener containing TCPA was introduced, although TCPA had been used in other hardeners since the plant opened. Production volume had approximately doubled during 1988 and in 1989 another TCPA-containing hardener was added to the process.

Local mechanical exhaust ventilation was provided for the soldering pots, hot stamping, and printing work stations. Installation of exhaust ventilation for molding presses was begun in July 1990 and completed by October 1990, after this survey was completed.

METHODS

Environmental assessment

Air sampling was performed in 1989 and 1990 for TCPA, TDI (in the inking/printing area), and formaldehyde, a pyrolysis product of colophony (in the coil winding/soldering area). Personal and area samples were collected for TCPA by drawing air at a flow rate of 1.0 L/min through 1.0 μm pore size Teflon filters connected to Tennax tubes (SKC Inc., Eighty Four, Pa.). The filter collected TCPA particulate, and the Tennax tubes collected the vapor phase of TCPA with analysis by gas chromatography. Samples for TDI were collected through two glass impingers connected in series, each containing nitro reagent (National Institute for Occupational Safety and Health Method No. MR 240) and were analyzed by high-performance liquid chromatography. Samples for formaldehyde were collected through glass impingers that contained deionized water and were analyzed colorimetrically.

Concentrations of TCPA on the day of the cross-sectional survey were measured as both personal and area exposures in all manufacturing and front office areas. For calculation of geometric mean personal TCPA concentrations and the correlation of personal TCPA exposure with intrashift change in FEV_1, concentrations recorded as less than the detection limit (DL) were taken as two thirds of DL as recently suggested by Waters et al.[7]

Exposure classification. Exposure was also classified in the following ways.

1. *Exposure group.* We established four exposure groups, listed by what was considered to be decreasing intensity of TCPA exposure: (1) molders ($N = 7$); (2) those workers ($N = 6$) who intermittently were involved with activities at or near the TCPA area (for example, electricians, maintenance workers, supervisors); (3) coil assembly workers ($N = 33$); and (4) office and engineering personnel ($N = 6$), who spent little time in TCPA areas.
2. *Duration of employment since 1988.* The new TCPA hardener was introduced in 1988.
3. *Cumulative TCPA consumption over time.* We obtained records of TCPA purchases in pounds by month since 1988 (Fig. 1).

Medical assessment

Index cases. The following summaries were obtained on the evaluations of two former workers referred to as index cases who were not employed at the time of the survey.

Case 1: A 33-year-old woman, hired August 17, 1987, worked at winding coils and soldering. In September 1987, productive cough, wheezing, breathlessness, and chest tightness associated with ocular itching and rhinorrhea developed in the woman. In February 1988, a workplace challenge test to colophony fumes was conducted. Baseline PC_{20} was 3.5 mg/ml. After one breath of fumes from solder containing rosin core flux, an immediate fall in FEV_1 of 33% from the baseline level was documented; the response was prolonged and gradually reversed over 8 hours. The PC_{20} was 0.125 mg/ml at 8 hours and indicative of airway hyperresponsiveness. This study confirmed occupational asthma caused by rosin core flux (colophony).

Case 2: A 21-year-old woman, hired July 14, 1989, was employed as a coil assembler about 10 feet from

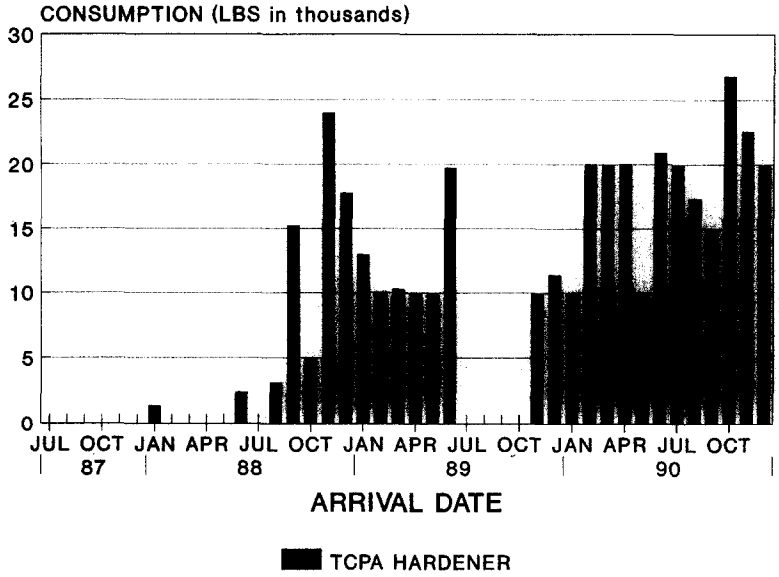

FIG. 1. TCPA product consumption by month of arrival.

molding. Cough, chest tightness, wheezing and dyspnea developed in this woman beginning in late September 1989, which became worse over the work week and improved on weekends. Within 1 hour on return to work on January 2, 1990, an acute asthmatic episode necessitated a visit to the emergency room. When the patient was seen on January 19, 1990, pulmonary function tests demonstrated a moderate nonobstructive ventilatory defect with decreases in all lung volumes: FEV_1 1.8 L (predicted 3.4), forced vital capacity (FVC) 2.1 L (3.8), total lung capacity 2.6 L (4.9). The diffusing capacity was decreased when corrected for lung volume. Chest x-ray film was normal with the exception of some bronchial wall thickening consistent with asthma. Methacholine test showed a PC_{20} of 6 mg/ml. Skin prick tests were positive to dust mite, ragweed, *Alternaria,* and cat and dog dander allergens. The serum TCPA-HSA-specific IgE level was elevated in January 1990 at an optical density (OD) of 0.44 (mean OD of nonexposed control subjects 0.016 ± 0.01); TCPA-HSA-specific IgG was 0.55 (mean OD of control subjects 0.03 ± 0.012). After the patient was treated with prednisone and cessation of workplace exposure, improvement in symptoms, FEV_1, and airway responsiveness (PC_{20} greater than 16 mg/ml) was seen; DL_{CO} was normal. This case was highly suggestive of IgE-mediated OA caused by respiratory sensitization to TCPA. A diagnosis of hypersensitivity pneumonitis was also entertained because of the diffusion and spirometry findings but was not supported by signs or radiographic evidence.

Initial survey. In July 1990 a cross-sectional evaluation was offered to all current employees, which consisted of an occupational and medical questionnaire and spirometry performed before and near the end of the work shift. The questionnaire was adapted from one used in several National Institute for Occupational Safety and Health Health Hazard Evaluations.[8] A chest symptom (wheeze, shortness of breath, or chest tightness) reported during the 3 months before the survey was considered work-related if (1) the onset was after starting work at the plant, (2) the subject reported temporal improvement (less frequently or not at all) on weekends or during vacations, and (3) it followed certain activities or exposures at work. Work-related asthma (WRA) was considered to be present by questionnaire if two or more work-related chest symptoms were reported.

For spirometric evaluation FVC, FEV_1, and FEV_1/FVC ratio were measured with a calibrated Vitalograph spirometer (Vitalograph, Buckingham, England).[9] At least three satisfactory expiratory maneuvers were performed; in all cases, the best two FVC and best two FEV_1 values agreed within 5% or 100 ml, whichever was greater. Percents predicted for FVC and FEV_1 were computed on the basis of the prediction equations of Crapo et al.[10] A 10% or greater decrease in FEV_1 across the work shift was considered a significant response.[11] It was not possible to incorporate into the protocol a test of airways reactivity or serial peak expiratory flow rate recordings.

Immunologic survey. Blood specimens were collected in October 1990 for serum specific IgE and IgG antibodies to TCPA-HSA. Because of turnover and absences, not all employees participated in both phases of the survey. Informed written consent was obtained from all subjects before blood specimens were obtained.

Preparation of chemical protein conjugates. TCPA was conjugated to HSA by methods previously described.[12] The moles of hapten on HSA were calculated as previously described by spectrophotometric analysis.[12] The final hapten-to-protein ratio was 13:1.

Specific IgG and IgE immunoassay. A modified

TABLE I. TCPA exposure data, Plant A, Ontario

Date	Location	No. of samples	Type of sample	Geometric mean TCPA concentration (mg/m³)	Range
Oct. 13, 1989	Preforming	1	Area	0.23	–
	Molding	3	Personal	0.21	0.14-0.34
	Molding	3	Area	0.32	0.17-0.59
May 1, 1990	Preforming	1	Area	0.29	–
	Molding	4	Personal	0.39	0.3-0.55
	Leak testing	1	Area	0.21	–
	Winding station	1	Area	0.28	–
	Crimping	1	Area	0.28	–
July 16, 1990	Preforming	1	Personal	0.13	–
	Preforming	1	Area	0.08	–
	Molding	5	Personal	0.0016	<0.009-0.05
	Molding	2	Area	0.011	<0.009-0.02
	Winding	1	Personal	0.009	–
	Soldering	1	Personal	0.009	–
	Finishing	2	Personal	0.008	<0.009-0.01
Nov. 23, 1990	Preforming	1	Area	0.03	–
	Molding	6	Personal	0.021	<0.01-0.11
	Molding	3	Area	0.009	<0.01-0.01
	Leak testing	1	Area	<0.01	–
	Winding	1	Area	0.01	–
	Front office	1	Area	<0.01	–

ELISA was performed to measure serum specific IgE and IgG to TCPA-HSA as previously described.[13] Immulon plates (Dynatech Laboratories, Alexandria, Va.) were coated overnight at 4° C with 10 μg/L of TCPA-HSA. After washing with phosphate-buffered saline–0.05% Tween (Sigma Chemical Co., St. Louis, Mo.), 100 μl of serum at dilutions of 1:50 and 1:100 was added and the plates were incubated at room temperature for 4 hours. To assay for a specific IgE, 100 μl of goat antihuman IgE antibody (Kirkegaard and Perry, Gaithersburg, Md.) was incubated in each well for 1 hour. After washing, rabbit antigoat IgE conjugated to alkaline phosphatase (Kirkegaard and Perry) at a 1:1000 dilution was incubated for 1 hour. After washing, p-nitrophenyl phosphate (Sigma Chemical Co.) prepared in diethylanolamine buffer (Aldrich Chemical Co., Milwaukee, Wis.) was added to each well. The OD was read on a Dynatech ELISA reader at 405 nm. The color reaction was terminated by addition of 2N NaOH when the positive control serum reached an OD of 0.6. The ELISA assay for specific IgG was performed similarly but as a four-step assay by sequentially adding TCPA-HSA, human sera, and goat antihuman IgG alkaline-phosphatase at 1:1000 dilution followed by the p-nitrophenyl phosphate substrate. Results greater than 3 SD above the mean OD of a control group not exposed to TCPA ($N = 12$) were considered positive.

Statistical analyses. The Statistical Analysis System (Version 6, SAS Institute Inc., Cary, N.C.) for personal computers was used for all analyses except for linear trend in proportions and Mantel-Haenszel procedures, for which True Epistat (Epistat Services, Richardson, Texas) was used. For categorical data, χ^2 and logistic regression were used, and for continuous data, correlation coefficients and linear regression were used. The duration of employment variable was not normally distributed; after log transformation, the distribution of durations did not differ significantly from a Normal distribution. In the logistic models for predictors of IgE positivity, only variables that produced a significant change ($p < 0.05$) by the likelihood ratio test (LRT) in univariate models were examined in further models.

RESULTS

Environmental assessment

TDI and formaldehyde (a marker for pyrolysis breakdown products of colophony) were undetectable when sampled on two previous occasions and on the day of the survey. Results of TCPA exposure data are listed in Table I. TCPA was detectable throughout the plant; on a given occasion, TCPA exposure did not vary markedly between nonoffice departments. Both TCPA vapor and particulate phases were detectable in approximately equal amounts. Exposures were considerably lower in July 1990. Consumption of the new TCPA hardener increased over time (Fig. 1).

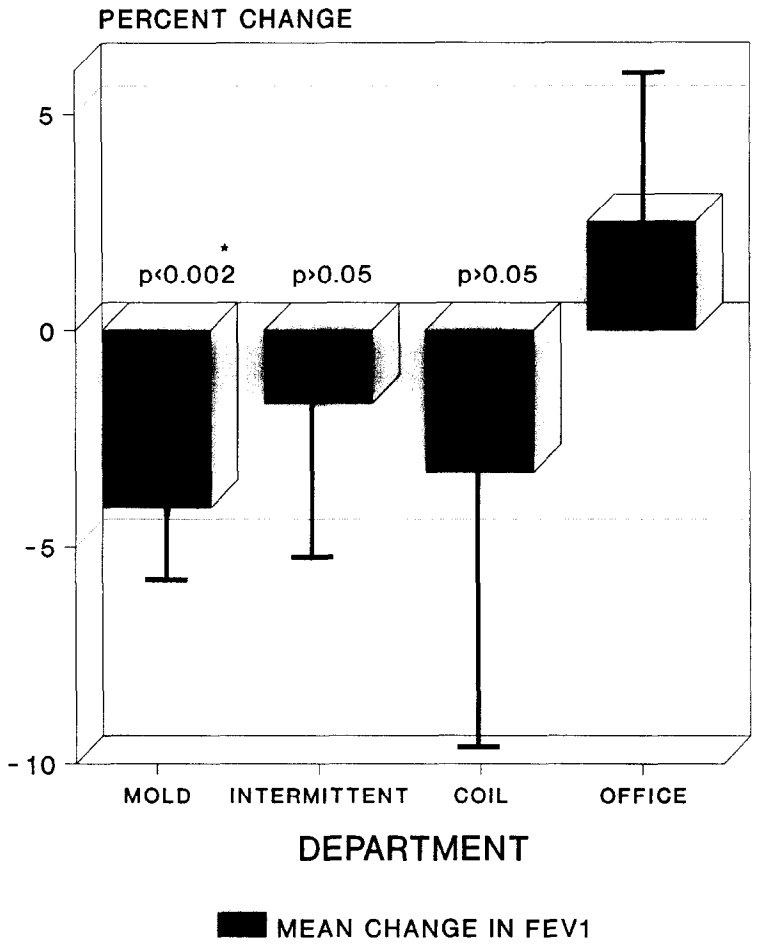

FIG. 2. Change in FEV_1 during shift, by department, Plant A, Ontario, July 1990. *Bars* represent SD. *Asterisk,* Probability in comparison with office, adjusted for current smoking.

Medical/epidemiologic assessment

Initial survey (July 1990)

Participants: Fifty-two (79%) of 66 current employees participated in the survey. Reasons for not participating included refusal (3 employees), absent or on holiday (3), compensation or injury (7), and maternity leave (1). The examined employees ranged in age from 16 to 67 years (mean 35.2) and had worked a mean of 2.5 years at the plant (range 0.1 to 8.1); 83% were women. All but six worked on the day shift (two in mold, three coil, one intermittent).

Symptoms reported: There was a high prevalence of reported respiratory symptoms on the questionnaire. As defined above, work-related wheeze, shortness of breath, and chest tightness were reported by 14 (27%), 15 (29%), and 20 (39%) subjects, respectively; WRA according to the above criteria was identified in 18 (35%). In general, wheeze, chest tightness, and WRA were reported most frequently by the mold workers.

Smoking and symptoms: Of the participants 27 (52%) were current smokers, 18 (35%) never had smoked, and 7 (14%) were ex-smokers. The proportion of current smokers was highest in the mold group. Wheeze and WRA were reported approximately twice as frequently in smokers and ex-smokers. After adjusting for smoking status, the differences between exposure groups in reported work-related symptoms were not significant (all p values > 0.1).

Spirometry. Only 2 (3.9%) of 52 participants had a baseline FEV_1/FVC ratio less than 70%; one was a mold worker and the other in the intermittently exposed group. One of these was a current smoker, the other an ex-smoker, with 28 and 37 pack-years of smoking, respectively. Baseline pulmonary function was lower in the ex-smokers and current smokers as expected; in linear regression analyses, pack-years was a significant predictor of breathing function (regression coefficient for pack-years was -0.018 ($p = 0.002$) for percent

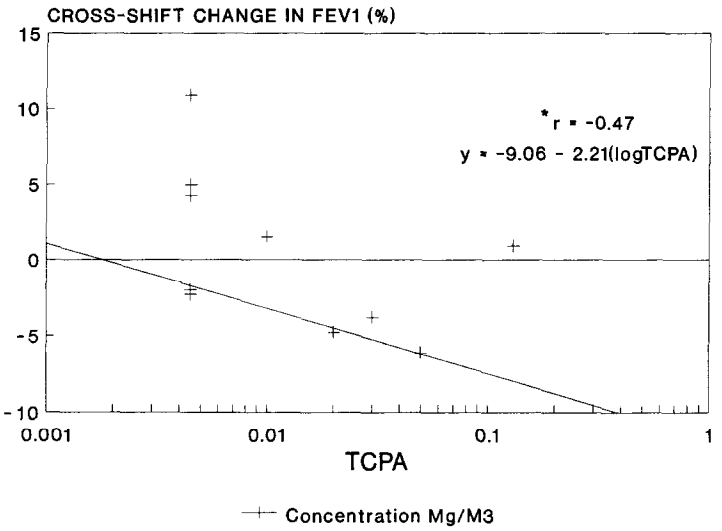

FIG. 3. Personal TCPA exposure versus cross-shift change in FEV_1, Plant A, Ontario, July 1990. *Asterisk,* Correlation with log (TCPA).

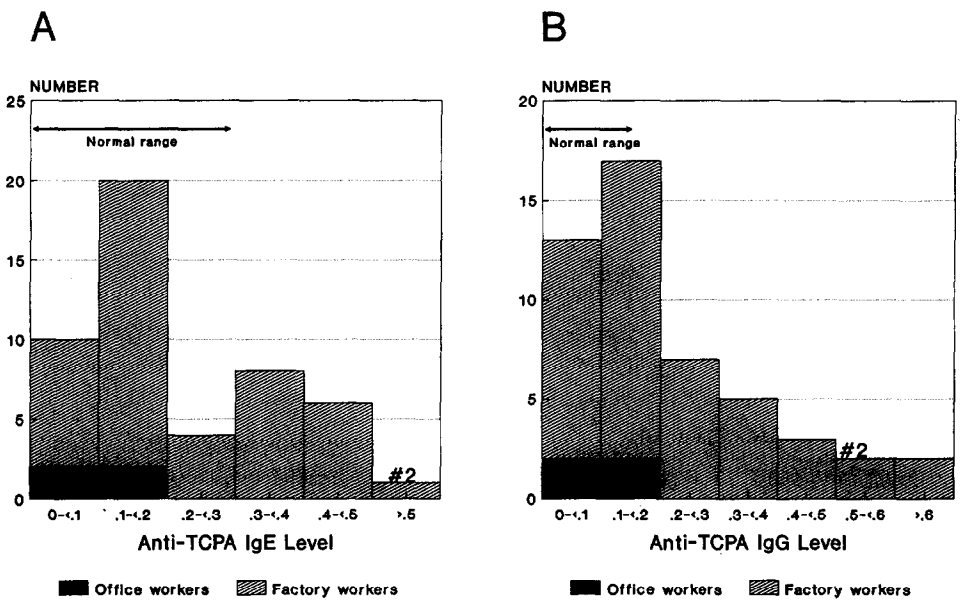

FIG. 4. Distribution of anti-TCPA-specific antibody levels expressed as OD: **A,** IgE antibody (serum dilution 1:50); **B,** IgG antibody (serum dilution 1:100). Among 12 nonexposed control subjects, the mean (±SD) levels were 0.09 (±0.07) and 0.05 (±0.05) for IgE and IgG, respectively. *#2,* Results for index case 2.

predicted FEV_1 and −0.013 ($p = 0.069$) for FEV_1/FVC). After adjusting for pack-years of smoking, there were no significant differences in baseline pulmonary function among exposure groups.

The differences among groups in mean change in FEV_1 during the shift were not significant by analysis of variance ($p = 0.2$). The average decline in FEV_1 (down 4.1%) was greatest in the mold group, and this was significantly different from results in the office group (2.5% increase; $p = 0.0016$ after adjusting for current smoking status; Fig. 2), whereas the change in the office group was not significantly different from that in the other groups ($p > 0.05$). There was a moderately strong negative correlation between intrashift change in FEV_1 and log TCPA concentration obtained from TCPA personal air samples in 10 workers ($r = -0.47$; Fig. 3); because of the small sample size, it was not significant ($p = 0.17$).

TABLE II. Relationship between exposure groups and antibody status, Plant A, Ontario, 1990

Antibody status	No. (% Positive)	Exposure group* 1	2	3	4	*p* Value	*p* Value (trend)†
IgE	15 (30)	3 (43)	4 (67)	8 (25)	0	0.093	0.019
IgG	19 (38)	4 (57)	4 (67)	11 (34)	0	0.124	0.022
Total samples	49	7	6	32	4		

*Exposure group 1 = evermold (in year before survey); 2 = utility/maintenance; 3 = coil assembly; 4 = office, not in plant. Includes all 49 subjects with antibody determinations.
†Exposure groups 1 and 2 combined for trend analysis.

TABLE III. Characteristics of those with antibody determinations, Plant A, Ontario, 1990

	IgE status among all those with serum samples			IgE status among those hired since 1988		
	Positive (*N* = 15)*	Negative (*N* = 30)*	*p* Value†	Positive (*N* = 12)*	Negative (*N* = 23)*	*p* Value†
Current smoking	3 (20)	17 (57)	0.02	3 (25)	11 (45)	0.123
Female	11 (73)	31 (91)	0.019	10 (83)	23 (100)	0.044
Age in years (mean ± SD)	40 ± 14	34 ± 8	0.033	37 ± 14	32 ± 8	0.130
Duration of employment: geometric mean log ± SD	1.9 0.65 ± 0.61	1.04 0.05 ± 1.36	0.826	1.5 0.41 ± 0.36	0.63 −0.46 ± 1.11	0.013
Duration of employment since 1988: geometric mean log ± SD	1.70 0.53 ± 0.42	0.90 −0.11 ± 1.17	0.044			
Mold or intermittent group	7 (47)	6 (20)	0.086	5 (42)	3 (13)	0.091

*Number in parenthesis is the percentage of subjects in that subset only.
†By χ^2 or Fisher's exact test except for variables with mean ± SD; then *t* test.

Seven (13.5%) of 52 subjects demonstrated a greater than 10% decline in FEV_1; six worked in coil assembly and had more direct exposure to colophony than other workers. Only one of the workers in groups 1 and 2, who had the greatest potential TCPA exposure, had such a decline.

Immunologic findings

Participants and proportion with specific antibody: Of the 49 available samples collected in October 1990, 15 (31%) were positive for TCPA-HSA-specific IgE antibody and 19 (39%) positive for specific IgG antibody (Fig. 4). Among the 40 subjects who participated in the July survey, the prevalences of elevated antibodies were 14 (35%) and 17 (43%) for TCPA-HSA-specific IgE and IgG, respectively. There was no relationship between antibody status and reported work-related symptoms, baseline spirometry, average decline in FEV_1 during the shift, or proportion with intrashift decline of FEV_1 greater than 10%.

Relation between specific antibodies and exposure group (all samples): Among all 49 samples, the prevalence of positive IgE (and IgG) antibody was highest in groups 1 and 2 with the most direct TCPA exposure and lower in the coil assembly group; no antibodies were detected in the small office group (Table II). There was a significant trend for an increasing proportion of workers having positive antibody status when they had "greater potential" for TCPA exposure (*p* value for trend 0.019 for IgE, groups 1 and 2 combined).

Characteristics associated with positive IgE antibody status: Because no subjects from group 4 (office) had TCPA-HSA-specific antibodies, analysis of characteristics associated with positive antibody status was limited to nonoffice workers

TABLE IV. Relationship between duration of employment and antibody status among those hired since 1988*, Plant A, Ontario, 1990

Antibody status	No. (% Positive)	Duration of employment (years)			p Value	p Value (trend)
		<0.33	0.33-1.33	>1.33		
IgE	12 (33)	0	4 (27)	8 (53)	0.067	0.022
IgG	15 (42)	1 (20)	5 (33)	9 (60)	0.181	0.074
Total no. of samples	35	5	15	15		

*Excluding those in group 4 (office).

(groups 1 through 3). As shown in Table III, those subjects positive for specific IgE antibody tended to be older, more likely to be male, and less likely to be current smokers. Among nonsmokers and ex-smokers combined there was a significantly higher proportion of subjects who had specific IgE antibody (12 [48%] of 25) than among current smokers (3 [15%] of 20). Duration of employment (on logarithmic scale) was not significantly different between those positive and negative, although duration of employment since 1988 (on log scale) was significantly longer among those who were antibody positive ($p = 0.044$). Only 4 (9%) of 45 subjects were atopic on the basis of questionnaire responses; none had specific IgE antibody.

There was a higher proportion of subjects who had IgE antibody who worked in the direct-exposure departments (mold and intermittent; 47%) than among those who were antibody negative (20%) (OR 3.5; $p = 0.086$, Fisher's exact test, two-tailed; prevalence odds ratio 2.35). Finally, we examined cumulative TCPA consumption during employment (on a plant-wide basis, not an individual basis, thus the result paralleled duration of employment). Cumulative consumption ranged from 0 to 316,338 pound-months (median 224,480). There was no significant difference between those who were IgE antibody positive and negative in mean TCPA consumption (240,036 ± 66,176 versus 177,507 ± 119,942 pound-months; $p = 0.07$) or log TCPA consumption (12.34 ± 0.32 versus 10.11 ± 5.00; $p = 0.09$). However, for 100% of the 15 subjects who were antibody positive, cumulative consumption exceeded 100,000 pound-months, whereas 19 (63%) of 30 who were antibody negative had consumption exceeding that level ($p = 0.008$, Fisher's exact test, two-tailed; odds ratio not calculable).

A separate analysis of predictors of antibody status was performed among the 35 subjects hired since 1988, which coincided with the introduction of the new hardener. Those positive for IgE were again somewhat older, all but two were women, and, as previously, 25% were current smokers compared with 45% of those without antibody ($p = 0.123$; Table III). Among those who were positive for antibody, 42% were in the mold or intermittent groups versus 13% of those who were antibody negative (OR 4.8; $p = 0.091$, Fisher's exact test, two-tailed; prevalence odds ratio 3.2). Mean duration of employment was significantly greater among those who were IgE positive ($p = 0.013$). Duration of employment was further categorized as follows: (1) less than 0.33 years (those hired after July 1990 when air sampling first demonstrated that TCPA exposure was low); (2) 0.33 to 1.33 years (employed for up to 1 year before lower exposures were demonstrated); and (3) more than 1.33 years (employed for more than 1 year before lower exposures were demonstrated).

As shown in Table IV, there was an increasing trend in the proportion of those with positive IgE (and IgG) antibody with increasing duration of employment category (p value for trend, 0.022). Compared with the prevalence among those employed less than 1.33 years, the prevalence of IgE positivity was more than twice that in those employed more than 1.33 years, in both current and noncurrent smokers (Mantel-Haenszel odds ratio controlling for smoking status 3.5; 95% confidence interval [CI] 0.56 to 23.88).

Multivariate analyses. Logistic regression models were used to examine the influence of possible predictor variables alone and simultaneously on specific IgE antibody status. Models that used log-transformed duration of employment provided the best fit for IgE positivity. Among all those who had antibody determinations, duration of employment (log scale) and older age (older than median versus younger) were not significant predictors in univariate models and being in the direct-exposure group was of borderline significance, whereas duration of employment since 1988 (log scale) and smoking status were significant. Both these latter two variables were also associated with significant change in the LRT

when included in the model together (χ^2_1 4.47 and 5.27, respectively). A term for the interaction between smoking and duration of employment since 1988 did not contribute significantly (χ^2_1 0.458; $p > 0.5$). In the final model predicting IgE status, which included both variables, the odds ratio for current smoking was 0.19 (95% CI 0.045, 0.80), and for log duration employed since 1988, the odds ratio was 2.39 (95% CI 0.93, 6.14) per 1 year. The regression coefficients and standard errors are based on the Wald χ^2 and thus differ slightly from the χ^2 values from the LRT.

In logistic regression models among those hired since 1988, only the duration of employment was a significant predictor of IgE positivity (χ^2_1 8.3, $p = 0.004$ on log scale; χ^2_1 6.4, $p = 0.011$ untransformed). Smoking status and older age alone did not improve the model significantly ($p > 0.1$ for both), and being in the direct-exposure department was of borderline significance. There was no interaction between smoking and duration of employment. The odds ratio for duration of employment (per 1 year) was 4.99 (95% CI 1.14 to 21.96); the ratio changed moderately when adjusted for department (4.05), or for department, smoking, and older age (3.39).

Analyses considering IgG antibody: The relation of TCPA-specific IgG status to smoking status was similar to that observed for IgE, with proportions positive among current smokers versus nonsmokers and ex-smokers of 25% versus 56% for all samples ($p = 0.036$) and 33% versus 50% for those hired after 1988, respectively. Duration of employment on a log scale did not differ between those who were IgG positive and negative but duration of employment since 1988 (log scale) was longer among those who were positive for antibody ($p = 0.067$). Among those hired since 1988, log duration was significantly longer among those who were positive for antibody ($p = 0.026$).

DISCUSSION

As noted, two clusters of OA caused by TCPA have been reported previously.[2-4] In this investigation, we have demonstrated specific IgE and IgG antibody against TCPA-HSA, as did Howe et al.[3] and Venables et al.[4] We found a high overall prevalence of IgE sensitization to TCPA in 31% of workers and a 53% prevalence among those employed for at least 1 year before July 1990. In contrast, Venables et al.[4] found specific IgE to TCPA-HSA by RAST among 8% of 300 factory workers. Differences in prevalences of serum specific IgE between these two facilities could be attributed to higher peak or average exposures at this plant or to features related to the process. In this regard, it may be important that TCPA vapor and dust were detectable in our study, whereas the exposure in the United Kingdom factory was thought to be to dust only.

Associations were found between the prevalence of TCPA-HSA specific IgE responses and both exposure status (based on department) and duration of employment since 1988. The fact that TCPA sensitization developed at an intermediate prevalence in workers in coil assembly who did not directly work with TCPA is not surprising for two reasons: first, this department was close to the molding area, and second, air sampling indicated exposure to TCPA throughout the plant.

Most cases of acid anhydride–induced asthma appear to develop after a variable latency period of exposure. In the study from the United Kingdom, cases of TCPA-induced OA developed within 2 years of the introduction of TCPA to the epoxy coating process.[1] All seven of the cases reported by Howe et al.[3] showed development of symptoms after employment for at least 1 year. In our study, the minimal interval after hire among those positive for anti-TCPA IgE was 6 months. Our finding of significantly longer duration of employment since 1988 among those who were IgE positive was also reported by Venables et al.[4] Our data suggest that the duration of exposure determines the risk for development of specific IgE responses to TCPA antigen.

In our survey, we found a high prevalence of what appeared to be work-related chest symptoms. In the absence of peak flow studies, variable airway obstruction could not be confirmed. The highest symptom prevalence and greatest mean decline in intrashift FEV_1 was in the mold group, which had the most direct exposure to TCPA. However, the group sizes were small, and in the absence of challenge studies, one cannot speculate about "causal agents" despite the compatible symptoms. It must be noted that other affected workers may have left the workplace without coming to medical attention. Cross-sectional studies, which are limited by survivor bias, may underestimate disease prevalence.[14]

Our findings with respect to the association of current smoking and anti-TCPA IgE are in contrast with data reported by Venables et al.,[4] who found a higher prevalence of specific IgE among current smokers. Although an association with smoking has also been found with other sensitizers such as platinum,[15, 16] it has not been found consistently. For example, smoking was as common among subjects with asthma who were exposed to phthalic anhydride as among subjects

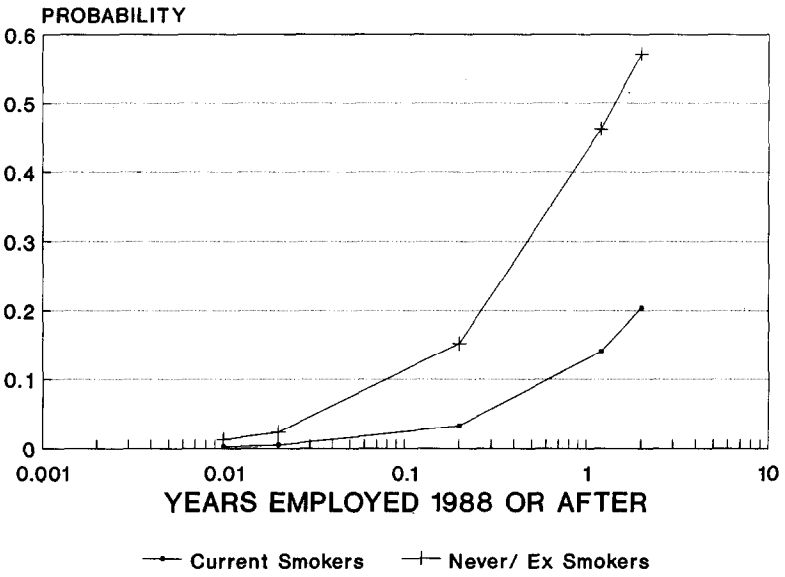

FIG. 5. Probability from logistic regression model of being anti-TCPA IgE positive, by years employed 1988 or after and smoking status.

who did not have asthma.[17] Seventy percent of workers with western red cedar asthma diagnosed by inhalation challenge were nonsmokers.[18] The explanation for the higher prevalence of sensitization among those who have never smoked and former smokers in our population and in other studies is not known. It is possible to speculate that a variety of changes in the microstructure of the airways induced by chronic smoking could modify the local immune response and thus either increase or decrease the potential for induction of specific IgE responses.

The logistic regression model suggested that both smoking status and duration of employment since 1988 were the most important predictors of IgE positivity. Among the subset of workers hired since 1988, only duration of employment (but not smoking status) was significant. Similar patterns were observed for predictors of IgG positivity. Considering all subjects, the model predicted that, for current smokers, the probability of being IgE positive if employed 0.2 years after 1988 was 0.033, increasing to 0.203 after 2.0 years; corresponding probabilities among nonsmokers and ex-smokers were 0.152 and 0.572, respectively (Fig. 5).

The reduction in TCPA exposure may have resulted in decreased sensitization among new workers. The fact that employees hired after July 1990 did not have IgE-mediated sensitization to TCPA could be attributed to reduction of TCPA exposure by means of new industrial hygiene measures or to an exposure period insufficient for sensitization. Prospective follow-up of this population will be required to address this question.

Different exposure measures. One can compare the relationship of possible parameters of exposures to outcome. Though measures such as job or department title, quantity of material consumed, or estimates of actual exposure may, in general, be preferable,[19] in this study duration of exposure as a surrogate measure appeared better, probably because of widespread contamination of TCPA throughout the plant.

In summary, although the rosin flux was likely responsible for some respiratory symptoms (positive colophony challenge was demonstrated in one index case and intrashift decreases greater than 10% clustered among coil workers), several pieces of evidence suggest a possible role for immunologic and respiratory sensitization to TCPA at the plant. First, there was a temporal association between the onset of breathing problems and the introduction and increasing use of new TCPA hardeners. Second, one index case of OA showed the presence elevated specific TCPA antibody. Third, there was a trend to higher prevalence of antibody positivity with duration of employment since the new TCPA hardeners were used. Fourth, the highest prevalence of antibody positivity occurred in the mold and intermittently exposed groups, which were most directly exposed to TCPA. Fifth, there was a negative correlation of cross-shift change in FEV_1 with personal TCPA exposure despite the low exposure levels. Sixth, air sampling did not detect isocyanates or pyrolysis products of colophony, and local exhaust ventilation was in place in recent years at the printing and soldering stations. Finally, there has

been a marked reduction in reported symptoms and no new cases of OA among newly hired workers at the plant since ventilation was installed and TCPA exposures have been documented to be lower.

On the other hand, those sensitized were neither more likely to report symptoms during the 3 months before the survey nor to demonstrate a greater decline in FEV_1 during the shift. Interpretation of these findings is difficult because the exposures documented at the time of the July survey were much lower than those measured at the plant previously. For example, TCPA exposures in May 1990 varied from about 3 to 20 times as high as those in July 1990. In fact, all but one TCPA air concentration measured in July was below 0.1 mg/m^3. It is noteworthy that Venables and Newman Taylor[6] reported "significant" changes in FEV_1 after TCPA challenge of subjects with proven TCPA-induced OA, at levels down to 0.1% TCPA (corresponding to 4 μg/m^3 TCPA in air). However, the mean difference in FEV_1 from baseline levels in their study exceeded 10% only at concentrations of 66.7 μg/m^3 (0.067 mg/m^3) or higher, or about 0.1 mg/m^3. Thus under the exposure conditions on the day of our survey, it would have been very difficult to demonstrate cross-shift declines in FEV_1 in this study, even if "true" cases of TCPA-induced OA were present.

The valuable assistance of Rita Edwards, Gary Ruffett, Frank Smith, and Peggy Doucette in facilitating the surveys is appreciated. We are indebted to the participation of Dr. Waldemar Maehle, nurses in the Hamilton office of the Ministry of Labour (MOL), and the staff of the MOL Chest Clinic in carrying out the spirometry and questionnaires. Ms. Irene Rule performed data computer entry and Ms. Marlene Vaz assisted with word processing. The Occupational Health Technologists in the MOL Hamilton office performed the air sampling. Dr. Fred Hargreave, McMaster University, kindly provided clinical notes on the index cases.

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Note added in proof. During our survey in 1990, a molder, hired in 1988, reported work-related wheeze and shortness of breath (and therefore satisfied our case definition for work-related asthma); she also had elevated levels of TCPA-HSA–specific IgE and IgG. After completion of this article, OA was confirmed in April 1993 on the basis of work-related changes in serial methacholine sensitivity and peak flow rate recordings.