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Respiratory Effects in Toluene Diisocyanate Manufacture: A Multidisciplinary Approach

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A new plant manufacturing toluene diisocyanate (TDI) has provided a unique opportunity to investigate the effects of TDI vapor inhalation on respiratory health in a group of exposed workers who have been studied prior to the start of plant operation. In order to establish dose–response relationships and determine host factors, complete biologic monitoring, including pulmonary function and immunologic studies, has been performed concurrently with a comprehensive environmental monitoring program including continuous sampling for atmospheric concentrations of TDI. Study groups include workers with regular exposure to TDI in production jobs (83), workers with intermittent contact with this vapor, usually in maintenance (28), and a control group of workers employed outside the TDI area (55). This population is being followed for a period of 5 yr.

The plant began operations in August 1973 with start-up procedures completed by the end of October. TDI spills occurred for numerous reasons, usually attributed to pump failure and resultant line blockage. Significant exposures also occurred in the drumming operation. The influence of these malfunctions is noted in the continuous monitoring data on atmospheric TDI concentrations which continue to reveal frequent excursions above the threshold limit value (TLV) of 0.02 ppm ceiling. These data are presented in relation to time and plant location.

Although the first full year follow-up following initial exposure was not complete, certain preliminary clinical observations were made. A number of workers had episodes of acute respiratory symptoms related to single exposure to an irritant gas at work, usually either TDI or phosgene. It appears that two or three workers in the study population have become "clinically sensitized" to TDI and have been removed from regular TDI exposure. To date, the total number of workers who report the presence of recurring respiratory symptoms has not increased in comparison with the pre-exposure survey.

Pulmonary function data after one full year of TDI exposure are not yet available. Pre- and post-shift ventilatory function studies do not indicate significant differences between the exposed and control groups. Selected individuals had carefully controlled inhalation challenge tests to monitored concentrations of TDI vapor under laboratory conditions. In workers suspected of having become "sensitized", immediate and/or late air flow obstruction was demonstrated and could be related to dose of inhaled TDI.

Although it has been recognized for over 20 years that an asthmatic or bronchitic response may be detected in workers exposed to toluene diisocyanate (TDI) vapor, either in manufacturing or polyurethane foam operations, comprehensive investigations designed to provide correlation of clinical, physiologic, immunologic and environmental data, have not thus far been attempted. A new plant manufacturing TDI has provided a unique opportunity to investigate the effects of TDI vapor inhalation on respiratory health in a group of exposed

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workers who have been studied prior to the start of plant operation. The objectives of this investigation are to characterize the pulmonary physiologic impairment of affected workers; determine the type of immune response and host factors associated with clinical manifestations; and describe the environmental exposure conditions in terms of TDI atmospheric concentrations at the job site. Factors to be considered in the investigation of occupational asthma include: environmental characterization, measurement of an adverse biologic response, mechanism of action, and host factors. As can be readily appreciated, a multidisciplinary approach is required in order to generate the component data. Industrial hygiene air sampling methods are used to characterize accurately the occupational environment of the worker. Modern methods for assessing the adverse biologic response of the lungs include not only the responses to an interviewer-administered questionnaire but also sophisticated pulmonary function techniques. Information regarding the inhalant environment and effect on respiratory health can be used to establish dose-response relationships as well as determination of a no-effect or threshold level for the atmospheric contaminant under study. The mechanism of action and possible pertinent host factors provide further information which is of obvious scientific importance as well as leading to the possibility of preventing the occupational disease, e.g., screening of potentially susceptible individuals. All of these techniques are being utilized in this study of workers exposed to TDI vapor in the manufacturing process.

Materials and Methods

Plant Environment

A new plant for the production of TDI was constructed in a pre-existing chemical manufacturing complex in Southwestern Louisiana. TDI operations began in August 1973, and start-up was considered complete by the end of October. The major areas within the TDI plant are: TDI synthesis and process loading; second-stage transfer or pumping station; finishing columns and TDI pump-out; and drumming. Since operations began, a number of TDI spills have occurred and these have not been limited to the start-up period. The spills have occurred for numerous reasons but are usually attributed to pump failure and resultant line blockage. Significant exposures have also occurred in the drumming operation.

Continuous 24-hr area sampling for TDI is being accomplished by use of UEI TDI detectors. These instruments continuously monitor atmospheric TDI concentrations up to 0.08 ppm. Sensitivity and specificity for TDI are obtained by utilizing a continuous reel of chemically impregnated paper tape supplied in cassettes which run for one week. In operation, air is drawn through the tape at a flow of 500 ml/min as it moves past an exposure orifice. If TDI is present, a stain is developed on the tape, the intensity being proportional to the concentration of TDI. The exposed tape then moves past two photo detectors, one measuring reflected light from the unexposed half. The two signals are then compared electronically and the output is proportional to the TDI concentration. The units have been calibrated in both the laboratory and under field conditions and correlate well with the Marcali method (Figs. 1 and 2). The detectors have been moved frequently so that all areas of the plant have been sampled.

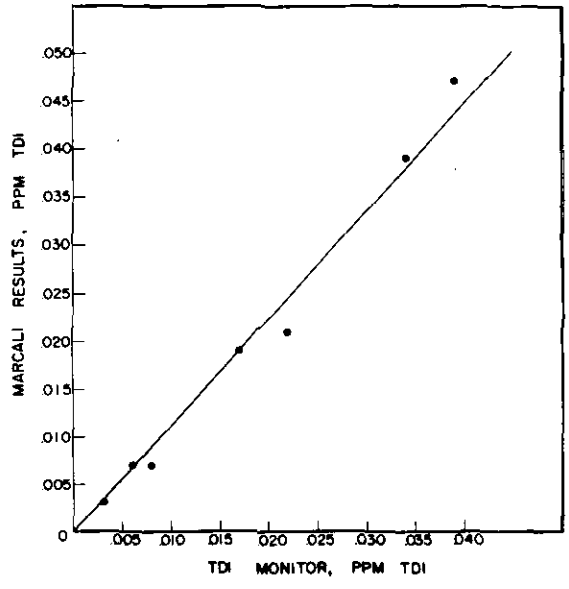

FIGURE 1. Laboratory calibration of TDI detector.

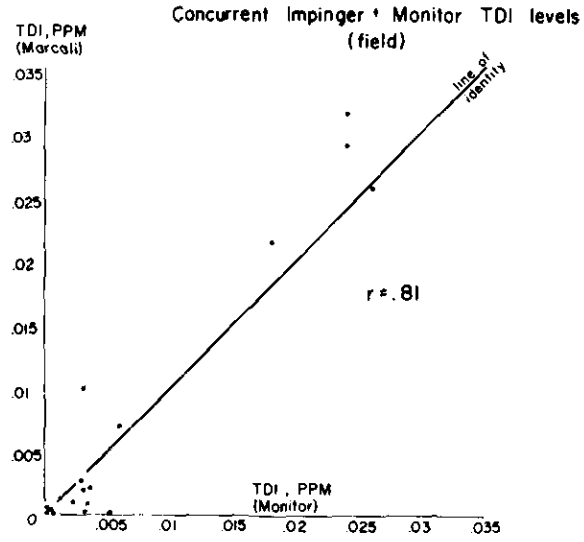

FIGURE 2. Field calibration of TDI detector.

Study Population

The population under investigation consists of three groups: workers engaged in production of TDI and who are more or less continuously at risk of TDI exposure (83), maintenance workers who have intermittent exposure to TDI (28), and employees of other chemical facilities within the complex who are presumed to have little or no TDI exposure and who serve as "controls" (55). The total study population of 166 was studied prior to start-up of TDI production and has been followed by clinical, pulmonary physiologic, and immunologic methods since the beginning of exposure.

Methods of Biologic Monitoring

Clinical

An initial questionnaire was administered by a trained interviewer using the National Heart and Lung Institute modification of the British MRC questionnaire with the addition of several questions adopted for this study. A follow-up questionnaire was designed by the investigators to detect change in respiratory health with the detection of any new or more prominent respiratory symptoms which may be associated with exposure at work.

Pulmonary Function

A completely self-contained mobile pulmonary unit was designed to be housed in a 27-ft Winnebago van and is fully operational. Complete studies of pulmonary function performed at the plant site include measurements of lung volume and compartments, maximum expiratory flow rates, ventilatory distribution, including closing volume, and pulmonary diffusing capacity. These measurements will be performed on a yearly basis and compared with the pre-exposure determinations. In addition, ventilatory function measurements are being performed every 4 months in order to provide early detection of any change in airways function. Recordings of both volume plotted against time (conventional spirometry) and maximum expiratory flow volume curves are being obtained with a separate but identical electronic spirometer, digitizing system, and rapidly responding X–Y recorder which is housed permanently at the plant site.

Immunologic

In April–May 1973, prior to commencement of TDI production, a visit was made to the plant to collect pre-exposure blood samples for eosinophil counts, immunoglobulin levels, and antibody assays, and to perform skin tests with common inhalant allergens and TDI–human serum albumin conjugates. A second visit was made in November–December, 1973, when blood samples were again collected following commencement of TDI production.

Serum was separated from the blood samples at the plant, frozen, and stored at —70°C until used to quantitate immunoglobulin levels (IgG, IgA, IgM, IgD, IgE), perform guinea pig PCA and monkey P-K tests, and detect anti-TDI antibodies by the radioimmunoassay and passive hemagglutination techniques.

Conjugation of TDI to Serum Albumin: TDI was conjugated to human and bovine serum albumin. In an ice bath, 100 ml of a 1% solution of the protein in 0.85% sodium chloride was adjusted to pH 8.5 with 1N NaOH. To this, 100 ml of 20 mg-% TDI in dioxane was added slowly over a 3-hr period, maintaining pH at 8.5 with 1N NaOH. The mixture was left in the ice bath for 1 hr and then transferred to a

dialysis bag and left in the hood overnight to allow dioxane to evaporate. Following dialysis in the cold against six changes of phosphate-buffered saline (pH 7.4), the conjugate was lyophilized and stored at —20°C until used. Conjugation was confirmed by ultraviolet absorption at 245 nm.

Skin Testing: A standard prick test was employed for immediate wheal and erythema responses. The common inhalant allergens used were *Aspergillus sp.*, Bermuda grass, house dust, *Alternaria sp.*, giant ragweed, pecan, oak, and plantain in 1:20 (*w/v*) concentration. For skin testing with TDI, 0.02 ml of a 5 mg per ml solution of HSA–TDI in saline was injected intradermally and the injection site observed for 30 min.

Eosinophil Counts: Eosinophils were counted in a Fuchs-Rosenthal counting chamber using a saponin-eosin stain and levels expressed as number of cells per cubic millimeter.

Immunoglobulin Quantitation: Immunoglobulins G, A, M, and D were quantitated by the single radial diffusion technique (*1*). IgE was quantitated by the radioimmunoassay technique described previously (*2*).

Anti-TDI Antibody Quantitation: Circulating anti-TDI antibodies were assayed for by two methods: radioimmunoassay by use of Sephadex G-25 superfine to which HSA–TDI antigen had been conjugated (*2*), and passive hemagglutination assay by use of sheep red blood cells to which HSA–TDI antigen had been conjugated by tannic acid (*3*),

In Vivo *Testing for Homocytotropic and Heterocytotropic Anti-TDI Antibodies:* Sera were tested for heterocytotropic anti-TDI antibodies by the passive cutaneous anaphylaxis technique (PCA) in guinea pigs (*4*) and for homocytotropic anti-TDI antibodies by the Prausnitz-Küstner method described by Layton (*5*).

Cellular Studies

Lymphocyte transformation was measured in selected individuals using a previously described method (*6*); lymphocytes from 50–60 ml of heparinized blood were diluted to give approximately 1×10^6 lymphocytes per tube. To appropriate tubes, 0.1 ml of antigen concentrations as used by Avery et al. (*7*) (0.5, 0.05, and 0.005 mg TDI–HSA per ml) was added. Following incubation in a CO_2 atmosphere 1 μCi (2 Ci/mmole) of thymidine was added to each tube, incubated overnight, and harvested. A trichloroacetic acid precipitate was placed into scintillation cocktail containing toluene, 1,4-bis[2-(4-methyl)5-phenoxyloxazoyl] benzene and 2,5-diphenyloxazole, and counted on a liquid scintillation counter. Results were recorded as DPM per tube.

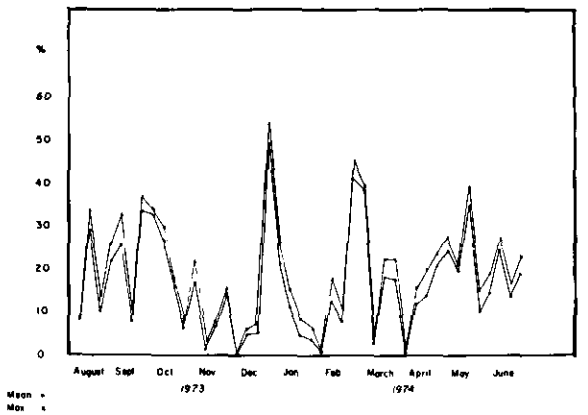

FIGURE 3. Per cent of sampling periods with (●) mean and (×) maximum TDI concentrations > 0.02 ppm (all locations).

Inhalation Challenge Testing

Selected workers who have apparently developed "clinical sensitivity" to TDI vapor exposure have been brought to the laboratory in order that carefully controlled and monitored exposure to TDI can be combined with repeated measurements of any adverse biologic response. After initial baseline evaluation, exposure was limited to 15 min, and in no case did the TDI atmospheric concentration exceed the threshold limit value (TLV) of 0.02 ppm. The worker was followed with appropriate monitoring until recovery from any adverse response.

Results

Environmental

The proportion of 15 min sampling periods in which the mean or maximum TDI concentrations exceeded the TLV of 0.02 ppm is shown (Fig. 3) and indicates frequent excursions above this present ceiling value. These

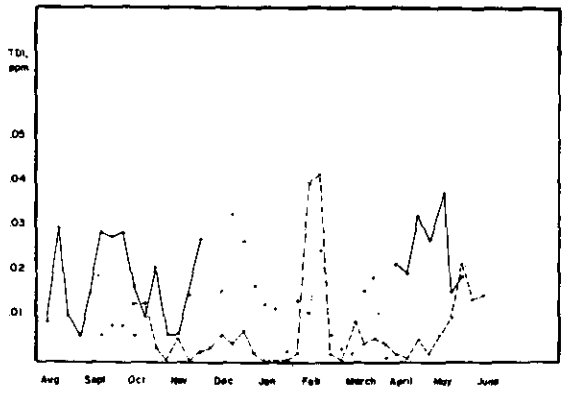

FIGURE 4. Average weekly maximum TDI concentration (by location): (. . .) TDI synthesis; (——) finishing and TDI pump-out; (--) drumming.

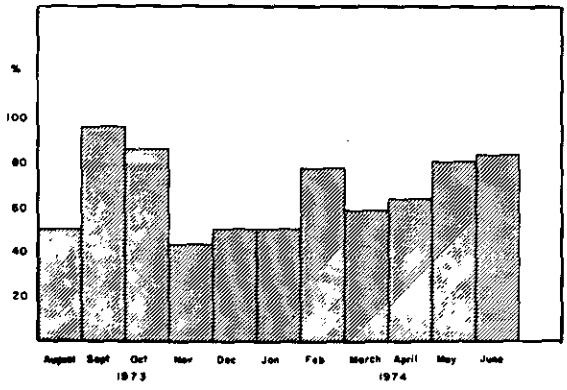

FIGURE 5. Per cent of days with average concentration TDI > 0.005 ppm (all locations).

excursions have occurred during the start-up, and have continued since the plant has been on line. The proportion of sampling periods with excessive concentrations has diminished overall since start-up was completed (Table 1), primarily because of improvement in the level of exposures in the drumming area. That all major locations have participated in excursions above the TLV can be appreciated by review of Figure 4. In view of the recently published NIOSH Criteria Document recommending an 8-hr time-weighted average of 0.005 ppm as the TDI standard, the proportions of days where the mean concentrations of TDI exceeded this level was plotted by month for the

Table 1. TDI mean and maximum concentrations in % of samples >0.02 ppm and (average in ppm) by plant location and time of operation.

Location	Before 11/1/73 (start-up)		After 11/1/73 (on-line)	
	Mean	Maximum	Mean	Maximum
TDI synthesis	3.2% (0.0056)	6% (0.0071)	15.1% (0.0113)	19.3% (0.0138)
Finishing	30.4% (0.0173)	33.3% (0.0205)	27% (0.0164)	33.3% (0.0211)
Drumming	13.5% (0.011)	16.1% (0.0129)	8.4% (0.0075)	9.6% (0.0088)
All	21.2% (0.0136)	24.3% (0.0165)	14.5% (0.0106)	17.5% (0.013)

plant as a whole. Although there was some monthly variation, in general the average TDI concentration exceeded the new recommended standard on approximately half of the days (Fig. 5). Considerable effort by the plant personnel is continuing in order that TDI exposures can be minimized so that average concentrations will be reduced.

Biologic Response

Clinical, physiologic, and immunologic data are markedly limited, since at the time of this report, the first full year of exposure has not yet elapsed. As of this time, a number of workers have presented with acute respiratory symptoms, generally short-lived, resulting from a single exposure to an irritant gas or vapor, usually phosgene or TDI. Most workers can accurately report which exposure had occurred, and often there is an associated recognizable malfunction as the source of his exposure. Ten to twelve workers appear to have become "clinically sensitive" to TDI vapor with such recurrent respiratory symptoms as cough, shortness of breath, chest tightness, and wheezing occurring after what seems to them to be minimal exposure to this vapor. These symptoms may have followed a heavy exposure to TDI initially but in some instances such a single episode did not seem to occur. These individuals have been removed from the TDI area to another part of the chemical manufacturing complex but not always without continuation of some of their respiratory complaints. The affected workers demonstrated a definite pattern of air

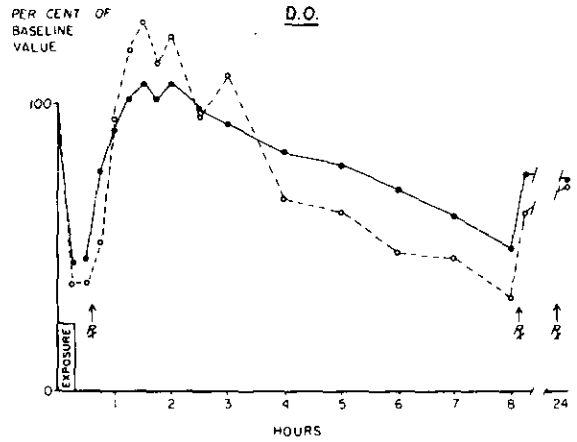

FIGURE 6. Inhalation challenge test: (●) FEV (1 sec); (O) FEF (25–75%). TDI = 0.0087 ppm. See text for details.

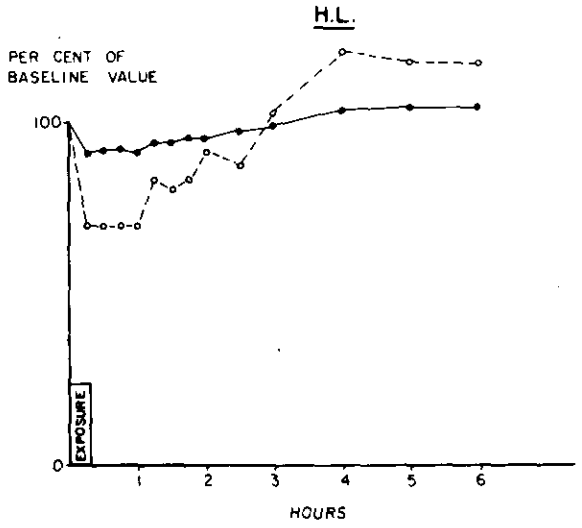

FIGURE 7. Inhalation challenge test: (●) FEV (1 sec); (O) FEF (25–75%). TDI = 0.0087 ppm. See text for details.

flow obstruction in association with their symptoms.

Inhalation challenge tests to TDI produced a variety of patterns in those clinically sensitized workers studied to date. A reduction in ventilatory function may occur both immediately following exposure and again several hours later, this late response being quite variable in degree and time required for recovery, the latter at times being longer than 24 hr. The characteristic dual response is shown in Figure 6. Another worker exhibited only a slight immediate reduction in ventilatory function

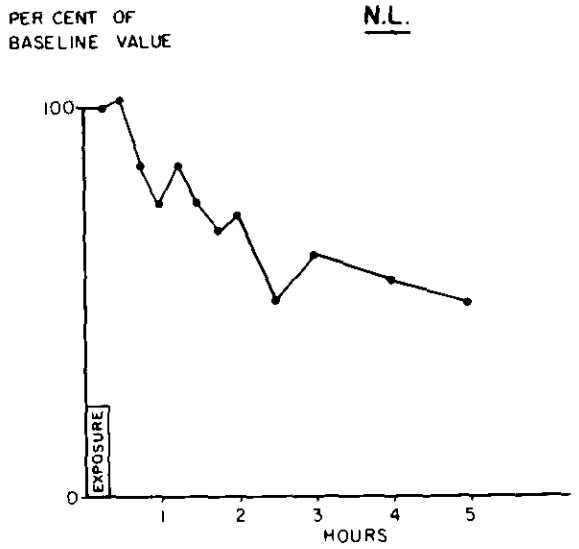

FIGURE 8. Inhalation challenge test: FEF (25–75%). TDI = 0.012 ppm. See text for details.

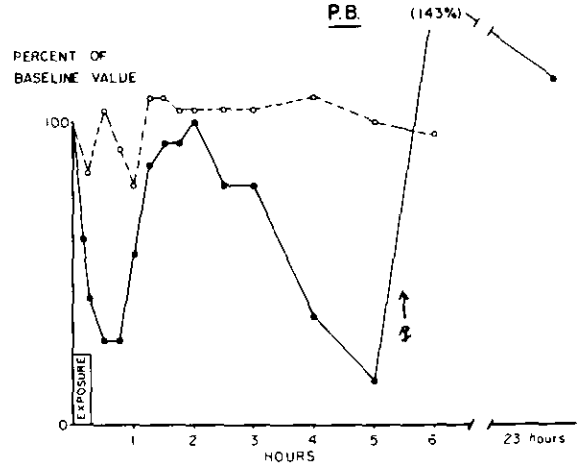

FIGURE 9. Inhalation challenge test: FEF (25–75%): (●) TDI = 0.0102 ppm; (O) TDI = 0.0028 ppm. See text for details.

and also demonstrated the greater sensitivity of the forced expiratory flow, 25–75%, in contrast to the more commonly measured forced expiratory volume, 1 sec (Fig. 7). The immeditate response may be absent entirely, with only a late response demonstrated following exposure (Fig. 8). It is obvious that in the plant setting, associating occupational exposure with respiratory symptoms may provide some difficulty in such an individual with a resulting delay in diagnosis. A dose–response rela-

tionship in a positive challenge test does not appear to have been previously demonstrated for TDI. In this study, a few individuals have failed to develop airways obstruction at one level of TDI but demonstrate the characteristic dual response at a higher level (Fig. 9). These early results do not yet establish the threshold level for TDI exposure, below which an adverse response will not occur, but hopefully additional data will be helpful in realizing this aim.

Information concerning a possible general long-term adverse effect of TDI on respiratory health must await at least the follow-up studies after one full year of exposure; definitive conclusions will not be possible until the completion of this multiyear investigation. Additionally, only preliminary data is available regarding host factors and mechanism of action associated with TDI respiratory effects.

On skin testing, 36% of group 1, 7% of group II, and 24% of group III subjects reacted to more than one of the eight inhalant allergens employed, indicating that each of the study groups contained atopic individuals. Positive reactions to HSA–TDI conjugate were observed in four subjects; however, testing with HSA alone gave similar reactions to those obtained with the HSA-TDI conjugate in each case, indicating lack of TDI specificity. Immunoglobulin quantitations showed a similar distribution for each group with no significant difference in mean values (Table I). Post-exposure samples showed no significant group difference with the exception of an unexpected increase of IgE levels in the nonexposure group from a mean value of 114.2 ng/ml to 635.3 ng/ml. Total eosinophil counts showed a similar distribution both before and after exposure, and there were no significant group differences.

PCA and P-K tests, performed with all post-exposure visit serum samples, were uniformly negative. The validity of these tests is not confirmed, however, due to a lack of known P-K and PCA positive control sera.

Passive hemagglutination and radiommunoassay (RIA) tests for specific TDI antibodies were negative in all study subjects. Although some sera yielded counts higher than background in the RIA, these were not sufficiently high to be considered definite positives (≥ 2 times background). RIA controls using serum from two rabbits immunized with TDI in complete Freud's adjuvant were positive, and tests with increasing amounts of these sera showed a linear relationship with counts per minute. Absorption studies indicated that the RIA technique was specific for anti-TDI antibodies.

Lymphocyte transformation tests were performed on four subjects who were "sensitized" to TDI and exhibited positive provocative bronchial challenge responses with aerosolized TDI in a controlled environment. No TDI–HSA-induced lymphocyte transformation was obtained above control values with any of the antigen concentrations employed; however, good stimulation was obtained with phytohemagglutinin in all subjects.

Comments

At the time of this preliminary report, a limited number of observations seem appropriate; the major body of information, which will hopefully ultimately result from this investigation, is yet to come. It is apparent that during the first year of operation in this plant significant periodic exposures to TDI vapor have occurred in a variety of operations; through the use of continuous monitoring, atmospheric TDI concentrations can be shown to exceed periodically but regularly the present TLV. Continuous area monitoring has added an important new dimension to the study of TDI respiratory health effects and the addition of continuous personal air sampling in the near future will provide even more valuable information concerning the individual worker's exposure to this vapor. The exposures to date have produced episodes of acute irritant effects with the appropriate respiratory symptoms, and also resulted in "clinical sensitivity" in several workers at current levels of TDI exposure. Carefully controlled inhalation challenge tests have provided confirmation of the adverse respiratory effect in these individuals and have demonstrated the varying patterns of response. While susceptibility to the effects of TDI has been suspected to be on an immunologic basis, these preliminary results indicate that the subjects tested to date, who worked in a TDI-containing environment, did not reveal a detectable immunologic response to TDI under the study conditions employed. Immunoglobulin levels did not change significantly following the start of TDI production with the exception of a marked increase of IgE levels in group III. The inability to detect IgE class-specific anti-

TDI antibodies during the early phase of study may indicate that a critical time–dose relationship exists between length and intensity of exposure to TDI. A further possibility is that TDI-induced bronchospasm may occur via a nonimmunologic mechanism such as nonspecific mediator release. It is obvious to both plant personnel and the investigators that more effective control of TDI vapor in the plant environment must be accomplished.

Acknowledgement

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