

CHRONIC TOXICITY SUMMARY

METHYL ISOCYANATE

(MIC, $CH_3-N=C=O$)

CAS Registry Number: 624-83-9

I. Chronic Toxicity Summary

<i>Inhalation reference exposure level</i>	1 $\mu\text{g}/\text{m}^3$ (0.5 ppb)
<i>Critical effects(s)</i>	Decreased weight gain and lung pathology at cessation of exposure in rats
<i>Hazard index target(s)</i>	Respiratory system; reproductive system

II. Chemical Property Summary (HSDB, 1995; CRC, 1994)

<i>Description</i>	Colorless liquid
<i>Molecular formula</i>	C_2H_3NO
<i>Molecular weight</i>	57.06 g/mol
<i>Boiling point</i>	39.5°C
<i>Melting point</i>	-45°C
<i>Vapor pressure</i>	348 torr @ 20°C, 600 torr @ 30°C (Varma and Guest, 1993)
<i>Solubility</i>	10 percent in water @ 15°C
<i>Conversion factor</i>	2.3 $\mu\text{g}/\text{m}^3$ per ppb at 25°C

III. Major Uses and Sources (Dave, 1985; U.S. EPA, 1986; HSDB, 1995)

Methylisocyanate (MIC) is prepared industrially by reacting methylamine with phosgene, oxidizing monomethylformamide at high temperatures ($\geq 550^\circ\text{C}$), or heating metal methylisocyanates. Because of its high reactivity, MIC is used as an intermediate in organic synthesis, most notably in the production of carbamate based pesticides. Tobacco smoke from some brands of cigarettes also contains MIC (about 4 μg per cigarette). Workers exposed to the MIC 8-hour threshold limit value of 0.02 ppm (46 $\mu\text{g}/\text{m}^3$) are exposed to approximately 460 μg MIC in a workday. Based on the most recent inventory, the annual statewide industrial emissions from facilities reporting under the Air Toxics Hot Spots Act in California were negligible (CARB, 2000). This does not include estimates of emissions of breakdown products from the use of metam sodium in agricultural applications. Use of metam sodium averaged 15,400,000 pounds/year from 1995 to 1999.

IV. Effects of Human Exposure

Although occupational exposures to MIC have been documented (Varma, 1986), few known exposures to the general public have occurred. A major exposure occurred in Bhopal, India in December 1984. Because of the sudden, short-term release (30-45 minutes), no measurements occurred, but the air concentration was estimated as 13 ppm (Dave, 1985) to 100 ppm (Varma, 1986).

The chemical identity of the ultimate toxicant has not been unequivocally determined and may consist of more than one chemical species. Although the chemistry of MIC suggests that hydrolysis to methylamine and dimethylurea is rapid, such hydrolysis in moist air is probably slow, and the reaction with photochemically produced hydroxyl radical is also slow (chemical $T_{1/2}$ about 3 months) (U.S. EPA, 1986). Brown *et al.* (1987) have shown that the alkylisocyanates (e.g., MIC) are relatively resistant (compared to the arylisocyanates) to hydrolysis in water. Hence, despite the high water reactivity of MIC, this compound could possibly persist in the environment for many days after an initial release.

Within 5 days of the initial exposure to MIC at Bhopal, more than 2,000 deaths occurred (Dave, 1985), while 4,000 more deaths were documented during the following decade (Lepkowski, 1994). The initial symptoms among the population living near the MIC plant were irritation and difficulty in breathing (Varma, 1986). Blindness occurred in more than 10,000 exposed persons but later resolved in most cases (Andersson *et al.*, 1990). The acute damage that led to death was mainly to the respiratory system, most likely pulmonary edema, bronchospasm, and electrolyte imbalance (Varma, 1986). However extrapulmonary damage, including tissue anoxia, gastrointestinal symptoms, and muscular weakness, were also observed (Dave, 1985). Within a year of the exposure, survivors continued to exhibit damage to the lung and eyes. Fibrosis of the lungs was seen in 30 percent of this group (Dave, 1985).

Reproductive toxicity was observed among women exposed to MIC in Bhopal. Varma (1987) reported 43 percent unsuccessful pregnancies among 865 women who were pregnant at the time of the MIC release. Among the live births, 14 percent of the infants died within 30 days, whereas a death rate of only 3 percent for the same interval was recorded 2 years prior to the release. Bhandari *et al.* (1990) reported increased spontaneous abortions and neonatal deaths among exposed women who were pregnant at the time of exposure compared to a control group in another city. In the latter study, stillbirths and congenital malformations were similar in the exposed and non-exposed groups.

Non-reproductive, non-pulmonary responses were evident in a group of exposed Bhopal residents, 3-years following exposure to the MIC vapors. Loss of vision and loss of visual acuity were more prominent among exposed residents than among unexposed people, and the losses appeared to be dose-dependent (Andersson *et al.*, 1990). In this study, the surrogate for dose was extent of early deaths in a housing cluster. Similarly, cataracts were reported more often among the exposed than among the unexposed group.

The lesions associated with lung damage may be expressed as pulmonary edema for immediate effects (Varma, 1986), and lesions associated with the bronchoalveolar area for long-term effects

(Dave, 1985, Varma, 1986). Vijayan *et al.* (1995) studied cellular components of bronchoalveolar lavage (BAL) and pulmonary function in Bhopal patients 1.3, 2.7, and 5.1 years after exposure to MIC. All had lived within 3-miles of the factory and all experienced acute respiratory and ophthalmic symptoms on the day of exposure. All were experiencing continued respiratory symptoms. Among the exposed people, decrements in forced vital capacity and forced expiratory volume (at 1-minute) were observed. In general, the decrements ranged from 12 - 21 percent of predicted values, whereas the control group exhibited decrements of 2 - 4 percent of the expected values. Analysis of the BAL revealed increases in total cells (all exposed groups), increased absolute numbers of macrophages (all exposure groups), decreased percentage of lymphocytes (2.7 and 5.1 year groups), and increased numbers and percentage of neutrophils (5.1 year group). These cell types are involved, through the secretion of various factors, in inflammatory and immunologic processes in the lung (Reiser and Last, 1986). The Vijayan *et al.* (1995) study thus suggests long term damage to lung parenchyma among people who survived the initial acute effects of MIC exposure.

In summary, humans exposed acutely by inhalation to MIC may experience long-term (as well as immediate) damage to pulmonary and extrapulmonary systems. The lung is probably the critical target organ for long-term effects from acute exposure, although adverse effects on other organs (e.g., eye, reproductive, and gastrointestinal) also exist. The late responses to the acute exposure suggest an immunological component, which could involve several systems including lung, eye, liver, and kidney. The chemical identity of the ultimate toxicant is unknown and may be more than one compound.

Avashia *et al.* (1996) assessed pulmonary effects from long-term, low-level MIC for more than 400 workers at a large chemical facility. Serial pulmonary function data, cigarette smoking histories, and industrial-hygiene measurements were available. Jobs were classified according to level of MIC exposure as none, low, moderate or high. Where work records were incomplete, exposures were based on the ratings of supervisors and coworkers. The frequency of pulmonary impairment was evaluated for the assumed four levels of exposure. No specific or consistent pulmonary impairment was evident. Unfortunately the report gave no quantitative classification of low, moderate or high MIC levels.

V. Effects of Animal Exposure

Experimental animal studies have been designed to address the experiences of the victims of the Bhopal disaster, in which the exposure has been described as acute because of the short duration (30-45 min). No studies were found that described exposure duration greater than 10 days. However, a chronic component to MIC exposure may exist as a result of slower rates of hydrolysis in air (compared to water), the presence of carbamylated hemoglobin in MIC-exposed people, and the change from edematous to inflammatory and/or fibrotic lesions with time. Further, a glutathione-dependent reversible MIC transport system has been suggested in experimental animals (see below).

MIC is absorbed through the respiratory tract and distributed to non-respiratory organs in experimental animals. In an acute (30 min) inhalation exposure to a dose of ^{14}C -MIC (labeled in

the isocyanate moiety) equivalent to one-LC₅₀ (23 mg/L), rats accumulated protein-bound radioactivity (including carbamylated proteins) in brain, liver, kidney, and lung, but not in blood (Bhattacharya *et al.*, 1988). Ferguson *et al.* (1988) exposed guinea pigs by inhalation to 0.47 ppm ¹⁴C-MIC (methyl group) for 6-hours. At the end of exposure, the label was found in arterial and venous blood, bile, and urine. At 2.7 days post-exposure, the label decreased to 2-7 percent. MIC was retained in the nasal-laryngeal area of the guinea pigs.

MIC, like reactive isocyanates in general, can react with biological molecules containing amino, alcohol, or sulfhydryl groups, as well as with water. While hydrolysis in an aqueous environment, such as the lung, is theoretically possible, measurements show that alkyl isocyanates are relatively resistant (compared to arylisocyanates) to such hydrolysis (Brown *et al.* 1987). The absence of a role for MIC hydrolytic products, methylamine (MA) or dimethylurea (DMU), is also suggested by the work of Jeevaratnam and Sriramachari (1994) and Sriramachari *et al.* (1994). Inhalation (30 min) or subcutaneous exposure of rats to either hydrolytic product at levels equivalent to the LC₅₀ or LD₅₀ did not result in death. Similarly, neither methylamine nor dimethylurea duplicated the acute effects of respiratory necrosis and congestion. However, exposure to these hydrolytic products did lead to interstitial pneumonitis, an observation that suggests MA and/or DMU could lead to subsequent inflammatory responses if sufficient amounts are present.

A role for methylamine in reproductive/developmental toxicity was investigated by Guest and Varma (1991). In a mouse study, pregnant dams were exposed to varying doses (intraperitoneal) of methylamine (as well as the di- and trimethyl compounds). Reproductive toxicity was not observed for methylamines. However, in cultured embryo experiments, decrements in crown-rump length, yolk-sac diameter, head length, and embryo survival were observed. The concentrations were high (>0.75 mM) and the interpretation of the biological activity of methylamine in terms of inhalation exposure is difficult.

MIC is a carbamylating intermediate; this is the basis for its use in the manufacture of carbamate based pesticides. In the same way, MIC should react with the appropriate functional groups of proteins, peptides, and nucleic acids. However, *in vitro* studies with cholinesterases show that such a reaction is not efficient (Brown *et al.*, 1987), an observation which may be explained by the presence of protonated amino groups at physiological pH (Baillie and Slatter, 1991).

A transport system for MIC via reduced glutathione (GSH) has been suggested by the discovery of the MIC-adduct, S-(N-methylcarbamoyl)glutathione (SMG), in the bile and the MIC-adduct of N-acetylcysteine (mercapturic acid, AMCC) in the urine of rats exposed to MIC by non-inhalation routes (Pearson *et al.*, 1990; Slatter *et al.*, 1991). The reaction of MIC with GSH and with cysteine is reversible, and can provide a source of free MIC in the tissues (Baillie and Slatter, 1991). Similar studies in experimental animals exposed to MIC by inhalation have not been reported. However, humans exposed by inhalation to N,N-dimethylformamide (H-C(=O)-N(CH₃)₂) excrete AMCC in urine (Mraz and Nohova, 1992). Hence a reversible MIC-transport system in animals, including humans, is possible, and the presence of high levels of GSH in human lavage fluid (Cantin *et al.*, 1987) would permit the initiation of this mechanism.

The toxicity of the adduct SMG was tested in mouse embryo culture (Guest *et al.*, 1992). Mouse embryos, at day 8 of gestation in vivo, were removed from their dams and cultured in the presence (and absence) of SMG. Dose-dependent (0.25 - 2 mM) decrements were observed for yolk sac diameter, crown-rump length, somite number, and protein content. Delayed DNA synthesis in the embryos and in yolk-sacs occurred in the presence of 0.25 mM SMG. Similar to the results obtained with methylamine, the SMG concentrations were high and the exposures were not by inhalation. However, the data show that a MIC metabolite, SMG, has toxic properties. In the presence of GSH (1 or 3 mM), the extent of the SMG-dependent toxicities was decreased. Such data demonstrate the reversibility of the binding between MIC and GSH.

Three inhalation studies were identified in which experimental animals were exposed to more than one dose of MIC. Among these studies, two used exposure durations for more than one day (Dodd and Fowler, 1986; Mitsumori *et al.*, 1987). Rats and mice were exposed by inhalation to 0, 1.1, and 2.8 (female) or 3.0 (male) ppm MIC for 6 hr/day for a total of 4 days, and then followed during a 91-day post-exposure interval (Mitsumori *et al.*, 1987). Among the rats, post-exposure deaths occurred by 49 days (male) and 14 days (female) at the high dose. Among the mice, only 1 male mouse died at 16 days post-exposure. Reduced weight gain was observed among the female and male rats in the high dose group, prior to death, although the absolute weights were not different from the unexposed rats one day before the end of exposure. Among the mice, a slowed weight gain was observed at 3- and 6-days post exposure (male) and 1 day post exposure (female) at the high dose, but normal weight gain returned by 1 week following cessation of exposure. At 7 days post-exposure, microscopic changes were observed in the respiratory system among the high dose rats of both sexes. Between 8- and 27 days post-exposure, increased lesions in the respiratory tract and also in liver, thymus, spleen, heart, and brain were observed at the high dose. Similar lesions were not observed in rats exposed to 1.1 ppm MIC and followed to the 8-27 day post-exposure. Among survivors, the incidence of lesions decreased to control values by 91 days. Among the mice, treatment related changes in the respiratory tract were observed at the high dose at 7 days post-exposure. Between 28 and 91 days, the lesions associated with the upper respiratory tract disappeared, whereas those associated with the major bronchi remained, although somewhat attenuated. These data suggest that the rat is more sensitive than the mouse to the effects of MIC. A LOAEL of 2.9 ppm is indicated, based on post-exposure decreased weight gain and respiratory tract changes in rats.

Dodd and Fowler (1986) exposed rats to 0, 0.15, 0.6, and 3.1 ppm MIC for two 4-day sessions at 6-hours/day and examined the animals within 1-day following exposure. The 2-cycle exposure included a 2-day recess from exposure. No deaths occurred at any MIC concentration during the exposure. Lesser weight gain occurred for rats in the 3.1 ppm groups, whereas weight among the rats in the 0.15 and 0.6 ppm MIC groups was indistinguishable from the air-exposed control animals. On exposure days 3 and 8, mean food consumption values in the high dose group were below those for the non-exposed group. At the time of termination, male rats exposed to 3.1 ppm MIC exhibited a 38 percent increase in hemoglobin concentration and a 26 percent decrease ($p < 0.001$) in oxygen saturation, compared to the unexposed rats ($p < 0.001$). Such changes were not observed for the female rats exposed to 3.1 ppm or for rats of either sex exposed to 0.15 or 0.6 ppm MIC. Absolute lung weights increased ($p < 0.001$) in both sexes after exposure to 3.1 ppm, compared to the control rats. Decreases in liver, kidney and testes absolute weights were observed in this exposure group, but the authors interpreted these data as a reflection of the body

weight losses. No weight changes were observed in rats exposed to 0.15 or 0.60 ppm MIC. Gross and microscopic lesions were observed in rats (female and male) exposed to 3.1 ppm, but not in rats exposed to 0, 0.15, or 0.6 ppm MIC. The microscopic lesions occurred in the respiratory tract and consisted of inflammation, epithelial necrosis, squamous metaplasia, and epithelial hyperplasia. These lesions extended into the bronchioles. These data suggest a NOAEL of 0.6 ppm MIC, based on weight gain loss, absolute lung weight, and lung histopathology in rats, immediately following cessation of exposure.

Post-exposure changes in lung pathology also occurred in the rats surviving 3.1 ppm in the Dodd and Fowler (1986) study. The early lesions associated with inflammation, epithelial necrosis, squamous metaplasia, and epithelial hyperplasia extending to the bronchioles either decreased in severity or receded toward the upper respiratory tract by 85-days post-exposure. In males, the intraluminal and submucosal fibroplasia changed in appearance during this interval, due in part to the maturation of fibrous tissue. Mucous plugs were also seen in the terminal bronchioles and alveoli in some rats. The importance of this observation is the progressive character of MIC induced lung disease. Such progression may be difficult to follow at lower doses, if the times involved are of insufficient duration.

Sethi *et al.* (1989) exposed rats by inhalation to 0, 0.21, 0.26, and 0.35 ppm MIC for 6 days at 0.5 hr/day. Statistical evaluation was not presented. No post-exposure deaths were reported, although lethality was recorded for rats exposed to 3.5 and 35 ppm for only 10 minutes. Following the 0.5 hr \times 6-day exposure, the weight gain declined in proportion to the exposure dose. At the lowest dose (0.21 ppm) the weight gain was 111 g after 91 days post-exposure, compared to a weight gain of 218 g during the same interval among the non-exposed rats. The absolute weights of the rats at the end of the exposure were not given. According to the narrative, inflammatory lesions of bronchopulmonary tissue were present; their extent increased with dose. A dose-response increase in markers of lung infection was present and suggests that the MIC exposed rats were more prone to infectious agents than were the unexposed animals. Non-specific lesions in liver and kidneys were also observed and appeared to be dose dependent, but the authors suggested that these effects could be a result of the lung infections.

Fetotoxicity was observed in two experimental animal studies (Schwetz *et al.*, 1987; Varma, 1987). Among female mice exposed to 0, 1, or 3 ppm MIC during gestation days 14 - 17 for 6 hr/day, an increased incidence of fetal deaths was observed at 1 ppm (Schwetz *et al.*, 1987). At 3 ppm, the average number of pups/litter decreased relative to the air-exposed controls. The dams were unaffected in terms of survival, body weight, or length of gestation. Non-gestational exposure (6 hr/day, 4 days) did not affect the number of pregnancies or the live litter sizes, suggesting that the fetotoxic effect may be specific to the female reproductive tract rather than a general attribute of systemic toxicity. Similarly, female mice exposed for 3 hours on gestation day 8 to 0, 2, 6, 9 or 15 ppm MIC gave birth to pups with decreased body weights at the lowest dose, although a good dose-response was not observed (Varma, 1987). At 9 or 15 ppm MIC, the surviving dams lost 75 - 80 percent of their fetuses. Maternal mortality and decreased skeletal lengths were also observed at 9 and 15 ppm. A distinction between maternally induced fetotoxicity and a direct effect on fetal health could not be made. Because the inhalation exposure to the dams occurred for only 3 hrs on one day, a chronic LOAEL is not suggested. Exposure of male rats to one dose of 3.2 mg/L for 8 minutes resulted in a 21 percent fertility rate

among the cohabited female rats within the day 8-14 period post-exposure compared to a fertility rate of 40% for controls; however, the rates increased after 15 days post-exposure (Agarwal and Bose, 1992). There was no evidence of fetotoxicity among the dams impregnated by the MIC-exposed male rats. Exposure of male and female mice to 0, 1, or 3 ppm MIC did not result in altered body weights, fertility, or litter size (Schwetz *et al.*, 1987). The results suggest that exposures to MIC at doses that are not toxic to adult male or female (pregestational) mice or rats do not result in adverse reproductive outcomes.

VI. Derivation of Chronic Reference Exposure Level (REL)

<i>Study</i>	Dodd and Fowler (1986)
<i>Study populations</i>	F344 rats
<i>Exposure method</i>	Inhalation (0, 0.15, 0.6, or 3.1 ppm)
<i>Critical effects</i>	Decreased weight gain and lung pathology immediately after cessation of exposure
<i>LOAEL</i>	3.1 ppm
<i>NOAEL</i>	0.6 ppm
<i>Exposure continuity</i>	6 hours/day, 8 days/10 day experiment (2-cycles, with one 2-day recess from exposure)
<i>Exposure duration</i>	10 days
<i>Average experimental exposure</i>	0.12 ppm for the NOAEL group (0.6 x 8/10 x 6/24)
<i>Human equivalent factor</i>	0.15 ppm for the NOAEL group (gas with pulmonary respiratory effects, RGDR = 1.23, based on BW = 152 g, MV = 0.12 L/min, SA = 225 cm ²)
<i>LOAEL uncertainty factor</i>	1
<i>Subchronic uncertainty factor</i>	10
<i>Interspecies uncertainty factor</i>	3
<i>Intraspecies uncertainty factor</i>	10
<i>Cumulative uncertainty factor</i>	300
<i>Inhalation reference exposure level</i>	0.5 ppb (1 µg/m ³)

Although the exposure was for only 10 days, the Dodd and Fowler (1986) study includes the longest exposure duration of the available investigations and also uses some of the lower exposure levels (down to 0.15 ppm). The microscopic findings of the respiratory tract were statistically analyzed, although an observation of the tabulated data at the four doses (0, 0.15, 0.6, and 3.1 ppm) clearly shows a NOAEL of 0.6 ppm. Other endpoints with the same NOAEL were increased hemoglobin and increased absolute lung weights. The symptomatic ramifications of the increased hemoglobin are unknown, although similar increases were reported for humans exposed to MIC in Bhopal (Srivastava *et al.*, 1988). The lung weight gain may be a reflection of the pathological changes seen in the microscopic studies.

Decreased body weight gain was also seen in the experimental 4 day rat inhalation study of Mitsumori *et al.* (1987) (NOAEL = 1.1 ppm), except that the decrease in the latter study did not

occur until 1 and 3 days (female and male, respectively) post-exposure. The apparent discrepancy could be explained, in part, on the basis of the length of exposure, which was twice as long in the Dodd and Fowler (1986) study. However, the weight gain loss in the Dodd and Fowler (1986) study was initiated within one day of the start of exposure.

The MIC chronic REL of 0.5 ppb is based on endpoints observed within 1 day of cessation of exposure. Post-exposure evaluation showed that, at a higher exposure level (3.1 ppm), progressive changes, including death, occurred. Post-exposure observations, however, were not reported at the 0.15 and 0.6 ppm MIC levels. The attribute of delayed MIC inhalation toxicity has also been observed in other experimental animals studies (Dodd and Fowler, 1986, Mitsumori *et al.*, 1987). In the case of the human MIC exposure in Bhopal, India, death did not occur during the immediate 30 - 45 minute exposure, but exhibited a lag phase. A few deaths occurred during the first few hours, the maximum occurred at 2 - 3 days, and by the end of a week about 2500 deaths were documented (Dave, 1985; Varma, 1986; Varma and Guest, 1993), although Varma (1986) suggests that the immediate number may be closer to 5,000. One report suggests that during the intervening decade as many as 6,000 deaths may be attributed to the initial exposure in Bhopal (Lepkowski, 1994). Such information suggests that the presence of an adverse effect at the NOAEL of 0.6 ppm (Dodd and Fowler, 1986) might be possible if the rats were observed during an extended post-exposure interval. Experimental evidence is needed to test this hypothesis.

Only one study was identified in which post-exposure observations were made on experimental animals exposed subchronically by inhalation to multiple doses of MIC. Mitsumori *et al.* (1987) exposed rats to 0, 1.1, and 2.8 (females) or 3.0 (males) ppm MIC for 6 hr/day for 4 days and observed the rats for 91 days. No deaths and no weight gain loss (in contrast to Dodd and Fowler, 1986) were present until the post-exposure period and were mainly observed in animals exposed at the high dose. Using a NOAEL of 1.1 ppm MIC, a chronic REL of 1.1 ppb ($2.6 \mu\text{g}/\text{m}^3$) was derived. The REL based on the Mitsumori *et al.* (1987) study is similar to the REL based on immediate effects (Dodd and Fowler, 1986), and may indicate that the time of occurrence of exposure related effects may not be as important as the MIC air concentration.

VII. Data Strengths and Limitations for Development of the REL

The strengths of the inhalation REL for MIC include the availability of controlled exposure inhalation studies in multiple species at multiple exposure concentrations and with adequate histopathological analysis and the observation of a NOAEL. Major areas of uncertainty are the lack of adequate human exposure data and the lack of chronic inhalation exposure studies.

VIII. Potential for Differential Impacts on Children's Health

Since exposures to MIC at levels that are not toxic to adult male or female (pregestational) mice or rats do not result in adverse reproductive outcomes, the chronic REL of $1 \mu\text{g}/\text{m}^3$ should adequately protect infants and children. MIC is a respiratory irritant and the developing respiratory system is more sensitive than that of adults. However, there is no direct evidence in

the literature to quantify a differential effect of MIC on the respiratory system of infants and children.

IX. References

Agarwal DK, and Bose M. 1992. Inhalation toxicity of methylisocyanate: assessment of germ cell mutagenicity and reproductive effects in rats. *Indian J. Exp. Biol.* 30:504-508.

Andersson N, Ajwani MK, Mahashabde S, Tiwari MK, Muir MK, Mehra V, Ashiru K, and Mackenzie DC. 1990. Delayed eye and other consequences from exposure to methyl isocyanate: 93% follow up of exposed and unexposed cohorts in Bhopal. *Br. J. Ind. Med.* 47:553-558.

Avashia B, Battigelli MC, Morgan WK and Reger RB. 1996. Effects of prolonged low exposure to methyl isocyanate. *J. Occup. Environ. Med.* 38(6):625-630.

Baillie TA, and Slatter JG. 1991. Glutathione: a vehicle for the transport of chemically reactive metabolites *in vivo*. *Acc. Chem. Res.* 24:264-270.

Bhandari NR, Syal AK, Kambo I, Nair A, Beohar V, Saxena NC, Dabke AT, Agarwal SS, and Saxena BN. 1990. Pregnancy outcome in women exposed to toxic gas at Bhopal. *Indian J. Med. Res [B]*. February. pp. 28-33.

Bhattacharya BK, Sharma SK, and Jaiswal DK. 1988. *In vivo* binding of [1-¹⁴C]methylisocyanate to various tissue proteins. *Biochem. Pharmacol.* 37:2489-2493.

Brown WE, Green AH, Cedel TE, and Cairns J. 1987. Biochemistry of protein-isocyanate interactions: a comparison of the effects of aryl vs. alkyl isocyanates. *Environ. Health Perspect.* 72:5-11.

CARB. 2000. California Air Resources Board. California Emissions Inventory Development and Reporting System (CEIDARS). Data from Data Base Year 1998. February 12, 2000.

Cantin AM, North SL, Hubbard RC, and Crystal RG. 1987. Normal alveolar epithelial lining fluid contains high levels of glutathione. *J. Appl. Physiol.* 63:152-157.

CRC. 1994. CRC Handbook of Chemistry and Physics, 75th edition. Lide DR, ed. Boca Raton, FL: CRC Press Inc.

Dave JM. 1985. The Bhopal methyl isocyanate (MIC) incident: an overview. In: Proceedings of an International Symposium, Highly Toxic Chemicals: Detection and Protection Methods. Schiefer, H.B. ed. Saskatoon, Saskatchewan, Canada. pp 1-38.

Dodd DE, and Fowler EH. 1986. Methyl isocyanate subchronic vapor inhalation studies with Fischer 344 rats. *Fundam. Appl. Toxicol.* 7:502-522.

Ferguson JS, Kennedy AL, Stock MF, Brown WE, and Alarie Y. 1988. Uptake and distribution of ^{14}C during and following exposure to [^{14}C] methyl isocyanate. *Toxicol. Appl. Pharmacol.* 94:104-117.

Guest I, Baille TA, and Varma DR. 1992. Toxicity of the methyl isocyanate metabolite S-(n-methylcarbamoyl)GSH on mouse embryos in culture. *Teratology* 46:61-67.

Guest I, and Varma DR. 1991. Developmental toxicity of methylamines in mice. *J. Toxicol. Environ. Health.* 32:319-330.

HSDB. 1995. Hazardous Substances Data Bank. TOMES® Vol. 25. Denver, CO: Micromedex, Inc. Expires 07/31/95.

Jeevaratnam K, and Sriramachari S. 1994. Comparative toxicity of methyl isocyanate and its hydrolytic derivatives in rats. I. Pulmonary histopathology in the acute phase. *Arch. Toxicol.* 69:39-44.

Lepkowski W. 1994. Ten years later - Bhopal. *Chemical and Engineering News.* 19 December 1994.

Mitsumori K, Boorman GA, Gupta BN, and Bucher JR. 1987. Four-day repeated inhalation and recovery study of methyl isocyanate in F344 rats and B6C3F1 mice. *Fundam. Appl. Toxicol.* 9:480-495.

Mraz J, and Nohova H. 1992. Absorption, metabolism and elimination of N,N-dimethylformamide in humans. *Int. Arch. Occup. Environ. Health.* 64:85-92.

Pearson PG, Slatter JG, Rashed MS, Han D-H, Grillo MP, and Baillie TA. 1990. S-(N-methylcarbamoyl)glutathione: a reactive S-linked metabolite of methyl isocyanate. *Biochem. Biophys. Res. Comm.* 166:245-250.

Reiser KM, and Last JA. 1986. Early cellular events in pulmonary fibrosis. *Exp. Lung Res.* 10:331-355.

Schwetz BA, Adkins B Jr, Harris M, Moorman M, and Sloane R. 1987. Methyl isocyanate: reproductive and developmental toxicology studies in mice. *Environ. Health Perspect.* 72:149-152.

Sethi N, Dayal R, and Singh RK. 1989. Acute and subacute toxicity study of inhaled methyl isocyanate in Charles Foster rats. *Ecotoxicol. Environ. Saf.* 18:68-74.

Slatter JG, Rashed MS, Pearson PG, Han D-H, and Baillie TA. 1991. Biotransformation of methyl isocyanate in the rat. Evidence of glutathione conjugation as a major pathway of metabolism and implications for isocyanate-mediated toxicities. *Chem. Res. Toxicol.* 4:157-161.

Sriramachari S, Rao GJ, Sharma VK, Jadhav RK, Saraf AK, and Chandra H. 1991. GC-NPD and GC-MS analysis of preserved tissue of Bhopal gas disaster: evidence of methyl carbamylation in post-mortem blood. *Med. Sci. Law.* 31:289-293.

Srivastava RC, Gupta BN, Athar M, Behari JR, Dwivedi RS, Hasan SK, Bharti RS, Singh A, Misra M, and Ray PK. 1988. Effect of exposure to toxic gas on the population of Bhopal: Part III - Assessment of toxic manifestations in humans - hematological and biochemical studies. *Ind. J. Exp. Biol.* 26:165-172.

U.S. EPA. 1986. United States Environmental Protection Agency. Health and environmental effects profile for methyl isocyanate. EPA/600/22. Environmental Criteria and Assessment Office, Office of Research and Development. Cincinnati OH: U.S. EPA.

Varma DR. 1986. Anatomy of the methyl isocyanate leak on Bhopal. In: Hazard Assessment of Chemicals. Saxena J, ed. New York: Hemisphere Publishing Corp. pp. 233-299.

Varma DR. 1987. Epidemiological and experimental studies on the effects of methyl isocyanate on the course of pregnancy. *Environ. Health Perspect.* 72:153-157.

Varma DR, and Guest I. 1993. The Bhopal accident and methyl isocyanate toxicity. *J. Toxicol. Environ. Health* 40:513-529.

Vijayan VK, Sankaran K, Sharma SK, and Misra NP. 1995. Chronic lung inflammation in victims of toxic gas leak at Bhopal. *Resp. Med.* 89:105-111