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**EVALUATION OF AIRBORNE EXPOSURE LIMITS FOR SULFUR MUSTARD:
OCCUPATIONAL AND GENERAL POPULATION EXPOSURE CRITERIA**

November 2000

**U.S. ARMY CENTER FOR HEALTH PROMOTION
AND PREVENTIVE MEDICINE**

**Aberdeen Proving Ground
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PREFACE

This report evaluates the occupational and general public air exposure limits for the chemical warfare agent sulfur mustard (HD) (CAS No. 505-60-2). It is one in a series of Airborne Health Criteria Documents on chemical warfare agents being prepared by the U.S. Army Center for Health Promotion and Preventive Medicine (USACHPPM) in conjunction with the Edgewood Chemical and Biological Center (ECBC) and other key organizations. The Airborne Health Criteria Document on the G-series nerve agents was finalized in April 1998 (ERDEC-TR-489), and that for VX nerve agent was finalized in February, 2000 (ECBC-TR-074). The scope and format of the nerve agent documents has been followed in the preparation of the current sulfur mustard analysis. The focus is on re-assessment of existing airborne exposure limits (for workers and the general population) by re-calculating those limits using methods that were not in existence at the time that the original limits were established. This document is primarily a health effects assessment that considers typical exposure scenarios for each population, and, as such, does not deal specifically with analytical detection capabilities, industrial hygiene and engineering controls, regulatory requirements, or unique exposure scenarios. All are important issues that must be considered by risk managers.

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EXECUTIVE SUMMARY

PURPOSE

The purpose of this document is to evaluate existing airborne exposure limits for occupational and general population exposures to sulfur mustard [bis(2-chloroethyl sulfide)] and, if necessary, to derive new exposure limits for both long-term and short-term exposures using the most current risk assessment methodologies.

DISCUSSION

Sulfur mustard (HD) is a vesicant chemical warfare agent capable of causing edema, inflammation, and necrosis of the epithelial tissues of the eyes, skin, and respiratory tract. Severe exposures can result in sufficient systemic uptake to cause gastrointestinal and hematological effects and immunosuppression. Exposures to sulfur mustard have also been associated with chronic bronchitis, recurrent keratitis, and cancers of the respiratory tract and skin.

Small quantities of sulfur mustard are used by various military and contract laboratories for defense research purposes, and verification of Chemical Weapons Convention compliance. Although bulk quantities are no longer manufactured in the United States, they currently exist in military stockpiles where they await eventual destruction. Quantities may also be found buried or abandoned at formerly utilized defense sites.

People whose work environment may include chemical weapon materials (whether in storage depots, demilitarization facilities, or research laboratories; as a consequence of treaty verification activities in support of the Chemical Weapons Convention; during remediation and/or decontamination of release areas; during emergency response operations, etc.) face potential risks of inadvertent exposure to this agent. To a much lesser degree, this risk is also shared by the general population in communities surrounding areas where chemical agents are stored, transported, or processed for disposal. In addition, chemical weapons, whether in foreign or domestic stockpiles, are still considered potential military threats and terrorist targets. The most likely route of exposure to sulfur mustard is by skin or eye contact, or by inhalation of the aerosol or vapor.

Existing airborne exposure limits (AELs) for sulfur mustard were promulgated by CDC (DHHS, 1988); AR 385-61 (DA, 1997a) and DA Pam 385-61 (DA, 1997b) also provide AELs for sulfur mustard. These AELs include an 8-hr/day, 5 day/wk TWA of 0.003 mg/m³ for occupational settings, as well as a 72-hr TWA of 0.0001 mg/m³ for the general population. The latter was intended for a 24 hr/day, 7 days/wk continuous exposure, and was published as a 72-hr TWA because of analytical limitations present at the time.

FINDINGS

General Population AEL for Chronic Exposures (GPL - General Population Limit)

The AEL for the general population (GPL) was calculated using both human and animal data. The available human data involved continuous exposures for a maximum time period of 600 min. Use of

short-term data requires the assumption of a linear response pattern over the time periods involved and may, to some degree, overestimate the potential effects if the response pattern is not linear (as suggested by the human studies). Even so, it was determined that a protective approach would be to use available human data to derive the AEL for the general population. The fact that the GPL derived from long-term animal data does not differ from that derived using human data supports the conclusion that the calculated GPLs are reasonable.

General Population AEL for Acute Exposures (AEGL - Acute Exposure Guideline Level)

While previous health criteria documents for other chemical warfare agents have included some proposed acute emergency guideline levels (AEGLs), the Army has recently realized that proper derivation of AEGLs involves a specific process including review and approval by a designated National Advisory Committee (NAC) for AEGLs. The Army is currently coordinating a separate effort with the NAC and now recalls any previously Army-proposed AEGLs. As such, no AEGLs are presented for sulfur mustard agent in the current document. The reader is referred to the *Federal Register* (65FR 14186-14197, March 15, 2000) for recent NAC-approved interim AEGL values for sulfur mustard agent.

Worker AEL for Chronic Exposures (WPL - Worker Population Limit)

As in the case of the GPL, the WPL for sulfur mustard was calculated using both short-term human exposure data and long-term animal data. The short-term human study involved three 8-hr exposures, one on each of three consecutive days. The effects seen under these test conditions were very mild symptoms of ocular toxicity. Since this exposure frequency is similar to that which workers would experience, the data are appropriate for calculating an 8-hr/day, 5 day/wk exposure limit. Although the same uncertainties exist in interpreting the results of this exposure in terms of possible cumulative effects following long-term exposures, Papirmeister et al. (1991) has stated that cumulative effects are less likely if the exposures are separated by a 2-3 day exposure-free period. Since workers would experience such a recovery period during weekends, the potential for cumulative effects may be greatly diminished. Nevertheless, additional uncertainty factors were used in deriving the WPL from the human data. The WPL derived from the human data is similar to that derived from the long-term animal data.

Worker AELs for Acute Exposures (STELs and IDLHs)

STEL (Short-term Exposure Limit). Human exposure data exist for calculating a STEL for sulfur mustard. A STEL (maximum of four 15-min exposures per day) calculated from the experimental data for single exposures resulted in values which, when averaged over an 8-hr work day, exceeded the 8-hr WPL. Another calculational approach, using a time-adjusted LOAEL, resulted in a value (0.0036 mg/m^3) very similar to the values calculated using probit analysis and logistics analysis (0.003 and 0.0067 mg/m^3 , respectively; with appropriate Uncertainty Factors). This comparison provides a degree of confidence that a STEL of 0.003 mg/m^3 is reasonable and protective. Further, the value of 0.003 mg/m^3 is a factor of 3 below the estimated no-effect concentration of 0.01 mg HD/m^3 for ocular effects. Therefore, for both technical and operational reasons, the recommended STEL for sulfur mustard agent is 0.003 mg/m^3 .

IDLH (Immediately Dangerous to Life or Health). Adequate human exposure data exist for calculating an IDLH for sulfur mustard. The data include exposure times of 30-33 min. At exposure concentrations of 0.06 to 1.7 mg/m^3 , the observed effects were no more severe than severe conjunctivitis.

The highest value of 1.7 mg/m³ was used to calculate an IDLH of 2.0 mg/m³. Although the data suggest that 30-min exposures to sulfur mustard air concentrations even higher than 2.0 mg/m³ may be below a true IDLH condition, the choice of 2.0 mg/m³ is considered to be appropriate in light of the possibility of increased sensitivity of workers who may have had previous exposures to the agent. Furthermore, because the data indicate the dose-response curve for sulfur mustard is relatively steep, i.e., 30-min exposures to 13.3 mg/m³ may cause severe eye damage and re-occurring keratitis years after the exposure, an IDLH of 2.0 mg/m³ would provide a greater margin of safety.

RECOMMENDATIONS

Based on the above discussions, this report's recommendations for sulfur mustard air exposure limits are as shown in the following table.

Existing and Recommended Airborne Exposure Limits for Sulfur Mustard					
Application	Type	Existing (mg/m³)	Recommended (mg/m³)	Exposure Time	Frequency
General population	GPL ^a (TWA) ^b	0.0001	0.00002	24 hr/day	7 days/wk, lifetime
Occupational	WPL ^c (TWA)	0.003	0.0004	8 hr/day	5 days/wk
	STEL ^d	NA	0.003	15 min	4 times/day
	IDLH ^e	NA	2.0	30 min	one time

^a GPL = General Population Limit (no observable adverse effects)

^b TWA = Time-weighted-average

^c WPL = Worker Population Limit; Occupational AEL (no observable adverse effects)

^d STEL = Short-term Exposure Limit

^e IDLH = Immediately Dangerous to Life or Health

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EVALUATION OF AIRBORNE EXPOSURE LIMITS FOR SULFUR MUSTARD

1. PURPOSE

The purpose of this document is to evaluate existing airborne exposure limits for occupational and general population exposures to sulfur mustard and, if necessary, to derive new exposure limits for both long-term and short-term exposures using the most current risk assessment methodologies.

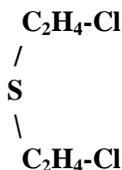
NOTE: While previous Criteria Documents for nerve agents (Mioduszewski et al 1998; Reutter et al 2000) have also included some proposed acute exposure guideline levels (AEGLs), the Army has recently realized that proper derivation of AEGLs involves a specific process of review and approval by a designated National Advisory Committee (NAC) for AEGLs. The Army is currently coordinating a separate effort with the NAC and now recalls any previous Army-proposed AEGLs. Therefore, no AEGLs are presented for sulfur mustard agent in this document. The reader is referred to the *Federal Register* (65FR 14186-14197, March 15, 2000) for NAC-approved interim AEGLs for sulfur mustard.

2. BACKGROUND

2.1 Introduction

Sulfur mustard (HD) is a vesicant chemical warfare agent capable of causing edema, ulceration, and necrosis of the epithelial tissues of the eyes, skin, and respiratory tract. Severe exposures can result in sufficient systemic uptake to cause gastrointestinal, hematological effects and immunosuppression. Exposures to sulfur mustard have also been associated with chronic bronchitis, recurrent keratitis, and cancers of the respiratory tract and skin. Information on adverse effects following long-term exposures to less-than acutely toxic concentrations is very limited. Health effects of sulfur mustard agents have been reviewed by NDRC (1946), NRC (1985; 1997), Papirmeister et al. (1991), Henry (1991), ATSDR (1992), Somani (1992), Sidell and Hurst (1992), Watson and Griffin (1992), Institute of Medicine (1993), Marrs et al. (1996), and Opresko et al. (1998).

Sulfur mustard is also known by the following chemical synonyms: *bis*(2-chloroethyl)sulfide; 1,1'-thiobis(2-chlorethane); 1-chloro-2-(2-chloroethylthio)ethane; distilled mustard; and agent HD. The Chemical Abstracts Service Registry Number for sulfur mustard is 505-60-2; its chemical formula is C₄H₈Cl₂S; and its chemical structure is:



2.2. Chemical and Physical Properties

Pure sulfur mustard (referred to as distilled mustard or HD) is a colorless, odorless, oily liquid with a molecular weight of 159.08 (MacNaughton and Brewer, 1994). Sulfur mustard found in munitions, however, often has a yellow-brown color due to contaminants (MacNaughton and Brewer, 1994) and a garlic or horseradish odor (DA, 1990). The physical and chemical properties of the pure agent are summarized in Table 1.

Table 1. Physical and Chemical Properties	
Molecular weight	159.08
Physical state	oily liquid
Boiling Point	217°C
Freezing Point	14°C
Solid density	1.37 gm/cm ³ at 0°C
Liquid density	1.27 gm/cm ³ at 20°C
Vapor density	5.4 (air = 1)
Vapor pressure	0.072 mm Hg at 20°C 0.11 mm Hg at 25°C
Volatility	75 mg/m ³ at 0°C (solid) 610 mg/m ³ at 20°C 920 mg/m ³ at 30°C
Henry's Law Constant	2.1 x 10 ⁻⁵ atm-m ³ /mol
Water solubility	0.68 g/L at 25°C 0.92 g/L at 22°C
Hydrolysis half-life	8.5 min in distilled water at 25°C
Octanol-Water Partition Coefficient (log K _{ow})	1.37
Soil Partition Coefficient (K _{oc})	133

SOURCES: DA, 1974, 1990; Small, 1984; MacNaughton and Brewer, 1994

The vapor pressure of 0.11 mm Hg at 25°C, indicates moderate volatility: a vapor concentration of 610 mg/m³ has been reported for a temperature of 20°C (DA, 1974); this is considered to be the saturation concentration above a pure liquid. The freezing point of sulfur mustard is 14°C; therefore, volatilization will be retarded under cold ambient conditions. Field data on air concentrations under various environmental conditions following spills or releases were not located in the available literature.

However, Rosenblatt et al. (1995) estimated the theoretical air concentration of sulfur mustard above a 10,000 m² parcel of soil contaminated with 1 mg of agent per kg of soil. For a worst case scenario, the average concentration over a 90-day period was estimated to be 0.0085 µg/m³. Rosenblatt et al. (1995) emphasized that empirical evidence as well as the measured reactivity of the agent would suggest that this value grossly exaggerated the potential exposure.

For the chemical weapons incinerator program, the U. S. Army estimated the air concentration of sulfur mustard at the boundary line of Aberdeen Proving Ground (DA, 1987). Air dispersion modeling indicated that the annual average concentration at the boundary line would range from 1.3 x 10⁻⁵ to 3.7 x 10⁻⁵ µg/m³.

2.3 Biological Properties

2.3.1 Mechanism of Action

The acute toxic effects of mustard vesicants are usually attributed to the consequences of alkylation reactions with organic compounds such as DNA. Alkylation reactions can result in physiological and metabolic disturbances as well as genotoxic effects.

As discussed by Papirmeister et al. (1991), the cytotoxic effects of sulfur mustard are dose-dependent, and are due, in part, to the fact that DNA is more sensitive to mustard-induced alkylation than other cellular constituents. Consequently, the low-dose effects of sulfur mustard, associated with 2 x 10² to 2 x 10⁴ alkylations per genome, are characterized by genotoxicity and inhibition of mitosis (Papirmeister et al., 1991). The mechanisms responsible for the loss of cellular reproduction are bifunctional alkylation reactions resulting in interstrand and possibly intrastrand DNA cross-links which prevent the separation of the complementary strands as required for normal DNA replication. Because energy metabolism and synthesis of RNA and protein are largely unaffected, such cells may undergo unbalanced metabolism and differentiation. Sulfur mustard-induced inhibition of cell division has been used to control hyperplasia of epithelial cells, *in vivo*, at dose levels of about 0.01 µg/cm², 10 to 100-fold lower than the dose (0.1-1.0 µg/cm²) causing erythema of the skin (Papirmeister et al., 1991). Similarly, in studies on rats Friedenwald et al. (1948) estimated that a dose of 0.01 µg/cm² is the threshold for inhibition of mitosis in corneal epithelial cells. Low-dose mutagenic (and possibly carcinogenic) effects are likely to be caused not by the bifunctional alkylation reactions inducing cross-links, but by monofunctional DNA damage.

At high doses the cytotoxic effects of sulfur mustard are associated with higher levels of alkylations per genome (2 x 10⁶ to 2 x 10⁷) resulting in depletion of nicotinamide adenine dinucleotide (NAD⁺), inhibition of glucose utilization, loss of plasma membrane integrity, and loss of normal cytoarchitecture (Papirmeister et al., 1991). For skin and corneal epithelial cells there is a good correlation between the amount of mustard fixed (i.e., that alkylates macromolecular cellular constituents and is not extractable) per unit surface area and the severity of the resulting lesion (Papirmeister et al., 1991). Friedenwald and Buschke (1948) reported that the death of corneal epithelial cells occurs at a sulfur mustard dose 10-20 times that causing inhibition of mitosis.

Several hypotheses have been advanced concerning the primary cause of cell death following acute exposures. As reviewed by Papirmeister et al. (1991), these are:

1. Poly(ADP-ribose) polymerase (PADPRP) hypothesis. - In this theory DNA is the initial target of the

- mustard agent. Alkylated DNA purines undergo spontaneous and enzymatic depurination, leading to the production of apurinic sites which are cleaved by apurinic endonucleases to yield DNA breaks. Accumulation of DNA breaks leads to activation of the chromosomal enzyme PADPRP, which utilizes NAD^+ as a substrate to ADP-ribosylate and a variety of nuclear proteins, causing severe lowering of cellular NAD^+ . Depletion of NAD^+ results in the inhibition of glycolysis, and stimulation of the nicotinamide adenine dinucleotide phosphate (NADP^+)-dependent hexose monophosphate shunt (HMS) pathway follows as a result of the accumulation of glucose-6-phosphate, a common precursor for both glycolysis and the HMS. Induction and secretion of proteases is stimulated as a result of enhanced HMS activity, and this leads to pathological changes in the cell.
2. Thiol- Ca^{+2} peroxidation hypothesis. The first step in this process is thought to be the alkylation of glutathione (GSH) by the mustard agent. Depletion of GSH subjects protein sulfhydryl groups to damage from the agent or from reactive cellular oxidants. Proteins most susceptible to damage include Ca^{2+} translocases (Ca^{2+} -stimulated, Mg^{2+} -dependent ATPase) which are dependent on thiol groups to maintain cellular Ca^{2+} homeostasis, and microfilamentous proteins, where loss of sulfhydryl groups could result in disruptions of the cytoskeletal and structural integrity of the plasma membrane.
 3. Lipid peroxidation hypothesis. According to this hypothesis the mustard agent causes depletion of GSH which, in turn leads to the buildup of highly toxic oxidants, usually through H_2O_2 -dependent reaction sequences. The oxidizing agents react with membrane phospholipids to form lipid peroxides, initiating a chain reaction of lipid peroxidation which can lead to alterations in membrane fluidity, loss of membrane protein function, and loss of membrane integrity.

2.3.2 Absorption, Distribution and Metabolism

Absorption. Because of its high lipophilicity, toxicologically significant amounts of the sulfur mustard are readily absorbed into epithelial tissues (Papirmeister et al., 1991). Most of the information on absorption rates is based on skin studies, and relatively little is known about absorption rates through the respiratory tract. Cameron et al. (1946) calculated the absorption of sulfur mustard vapors in the noses of rabbits and rhesus monkeys. The nose concentration of the agent was 10-30% of the chamber concentrations (40, 100 and 500 mg/m^3), suggesting a 70-90% absorption rate.

Absorption into the skin is dependent on the thickness of the epidermis and on the presence of moisture, which enhances penetration. Absorption may also be higher at the base of hair shafts and in the hair follicle where the epithelial tissue is thinner than the surrounding surface area (Papirmeister et al., 1991). Of the amount of sulfur mustard contacting the skin, 80% evaporates and 20% is absorbed. Of the latter fraction about 12% remains at the site and the remaining 88% enters the circulation (Renshaw, 1946). Renshaw (1946) reported that the rate of penetration is 1-4 $\mu\text{g}/\text{cm}^2/\text{min}$ at a temperature of 75°F.

Riviere et al. (1995) reported that following application of 400 μg of radiolabelled sulfur mustard per cm^2 of isolated perfused porcine skin, penetration rates over 2-8 hr ranged from 2.9 to 6.7% and rates of absorption from 1.2 to 4.0%. The mean total recovery of the radiolabel was 9.3% (range 3.8-17.7%) indicating a substantial loss due to volatilization.

Distribution. Several studies using radiolabelled sulfur mustard have been conducted to determine the tissue distribution of the agent and its metabolites following percutaneous or intravenous

exposures. Young et al. (1944) reported that 15 min after rats were exposed percutaneously, radioactivity was found in all examined tissues except the eyes. Clemedson et al. (1963) reported a fairly even distribution of radioactivity in mice after either percutaneous or intravenous exposures, with the highest accumulations occurring in the nasal region, followed by the kidneys, liver and intestine. Bournsnel et al. (1946) reported maximum levels of radioactivity in the kidney, lungs and liver of rabbits following intravenous dosing. Studies conducted by Hambrook et al. (1993) on the uptake and distribution of radiolabelled sulfur mustard in the skin and blood of rats after cutaneous application indicate that much of the agent that enters the blood becomes bound to red cell constituents such as hemoglobin. Binding may also occur with glutathione which is present in relatively high concentrations in red blood cells.

Axelrod and Hamilton (1947) reported that 5 min after the eyes of rabbits were exposed to sulfur mustard vapor, the agent was concentrated in the cornea with lesser amounts in the iris, lens, and conjunctiva.

Metabolism. Several studies using radiolabelled sulfur mustard have evaluated the biotransformation of sulfur mustard after intravenous or intraperitoneal injection in rats. Davison et al. (1961) reported that after intravenous injection the major urinary product was glutathione-bis-chloroethyl sulfide conjugates (45% of total urinary radioactivity) and smaller amounts of sulfone conjugates (7%) and thiodiglycol and its conjugates (14.4%). Roberts and Warwick (1963) reported that after intraperitoneal injection the major urinary product was cysteine-bis-(β -chloroethyl)sulfone). Studies on humans suggest that thiodiglycol may be present in the urine for one week or more after exposure (Wils et al., 1988). Based on the available data, Papirmeister et al. (1991) concluded that hydrolysis to thiodiglycol and reaction with glutathione are the most important routes of detoxification.

2.3.3 Local vs Systemic Effects

Exposure to sulfur mustard can result in local and/or systemic effects depending on the extent and duration of exposure. Immediate systemic effects occur only at high exposures, but are invariably accompanied by severe local effects at the point of contact (Papirmeister et al., 1991). Local effects, however, can occur at exposure levels much lower than those producing systemic effects (Papirmeister et al., 1991). For exposures to sulfur mustard vapors, the eyes, skin and respiratory tract are most susceptible to damage. The eyes are considered more sensitive than the respiratory tract, which is more sensitive than the skin (Papirmeister et al., 1991; IOM, 1993; Barkley, 1999). The relative sensitivities of these tissues to sulfur mustard vapors is indicated by the cumulative exposures (Ct, the product of the concentration, in mg/m^3 and the exposure time, in minutes) producing a similar degree of injury (Table 2). The Ct is used as a measure of exposure because the severity of the effect is a function of both the concentration and the exposure time; however, the Ct required to produce a given effect may vary with the exposure duration, frequency of exposure and individual sensitivity. For multiple exposures that occur within 12 hr or less, the effects are likely to be cumulative, and multiple exposures, each below the threshold, can lead to injury (Papirmeister et al., 1991). For exposures spaced over a 2-3 day interval, the effect is likely to be less than that produced by the same Ct administered in a single day; thus, the lesions caused by a single exposure Ct of $100 \text{ mg}\cdot\text{min}/\text{m}^3$ were reported to be similar to those from a Ct of $300 \text{ mg}\cdot\text{min}/\text{m}^3$ given in four separate exposures over 6-12 days (Papirmeister et al., 1991).

For all exposure routes, there is a latent period between the time of exposure and the onset of effects. Latency can vary from several hours to several days. Generally, the latent period decreases with increase in the Ct. Latency for effects on the eyes is generally shorter than that for effects on either the skin or the respiratory tract (Papirmeister et al., 1991). Ambient temperature affects the latent period at a constant agent concentration in that cold temperatures cause effects to appear at higher Cts, and high temperatures (especially when combined with high humidity) cause effects to appear at lower Cts (Barkley, 1999).

Table 2. Ct (mg-min/m³) Endpoints for Sulfur Mustard Vapor Exposures in Humans

Organ	Threshold ^a	Injury (non-disabling)	Incapacitation (IC ₅₀) ^b	Permanent injury or death	References
Eye	12 (16-27°C) 2 (≥32°C) 3-10	50-100 40-80	200	>800	Gates and Moore, 1946; McNamara et al., 1975; Papirmeister et al., 1991; PCS, 1946; Stepanov and Popov, 1962; Urbanetti, 1988; DA, 1974; DA, 1990; DA/DAF, 1975; Barkley, 1999
Respir. Tract	12-70	<100	200	1000 1,500 ^e	Ganas, 1969; McNamara et al., 1975; PCS, 1946; Robinson, 1967; Sidell, 1990; Stepanov and Popov, 1962; Stroykov, 1970; DA, 1974; DA, 1990
Skin	50-200 (moderate temp) 25-50 (high temp)	100-300	1000 ^c 2000 ^d	10,000 ^e	Gates and Moore, 1946; McNamara et al., 1975; Papirmeister et al., 1991; PCS, 1946; Sidell and Hurst, 1992; DA, 1974, DA, 1990; NRC, 1997

SOURCES: IOM, 1993, Table 8-1, as adapted from Papirmeister et al., 1991 and Watson and Griffin, 1992; Barkley, 1999

^a Threshold corresponds to first indication of non-disabling signs

^b The Ct expected to incapacitate 50% of those exposed; incapacitation defined as inability to perform designated duties (IOM, 1993)

^c Temperature 32°C; low humidity

^d Temperature 21-27°C; high humidity

^e The LC₅₀; the Ct expected to cause death in 50% of those exposed

2.3.4 Acute Toxicity

2.3.4.1 Effects on the Eyes

Signs and Symptoms. Depending on the vapor concentration, exposure to sulfur mustard can result in ocular irritation, redness, lacrimation, burning pain, mild to severe conjunctivitis, swelling of the eye lids, photophobia, blepharospasm, and corneal damage (Papirmeister et al., 1991; Somani, 1992; IOM, 1993; Barkley, 1999). Corneal injury is characterized by edema, clouding, necrosis, infiltration of polymorphonuclear neutrophils, pannus development (vascularization and connective tissue infiltration beneath the corneal epithelium), and corneal opacity. Normal corneal epithelial regeneration can occur rapidly if the underlying stroma is intact, but if it is damaged, regeneration is incomplete with

recurrent erosion and vascularization (Somani, 1992). Exposure of the eye to liquid droplets of sulfur mustard can result in the rapid appearance of symptoms and severe corneal damage, with possible perforation of the cornea and loss of the eye.

According to Papirmeister et al. (1991), there are no known biological or physical factors, other than the vapor or liquid state of the agent, which exacerbate the ocular effects of the agent; ambient temperature and humidity do not alter the severity of the response. Barkley (1999) and his colleagues consider that colder temperatures will reduce the severity of effects at a given Ct. After severe exposures to sulfur mustard, recurrent keratitis and corneal ulceration can occur years after the initial exposure (IOM, 1993; see also Medema, 1986; Grant, 1986). A more detailed discussion of this phenomenon is provided in Section 2.3.7.

Studies cited in IOM (1993) indicate that severe cases of conjunctivitis occurring after exposure to Cts of 50-100 mg-min/m³ healed in 2-14 days. At lower Cts, the conjunctivitis cleared in several hours to several days. According to IOM (1993), prolonged intractable conjunctivitis occurs only after chronic exposures to sulfur mustard.

Exposure-Response Data. Observed Ct-related ocular effects of sulfur mustard, as summarized by Papirmeister et al. (1991) as well as Barkley (1999) and his colleagues, are presented in Table 3. Mild eye irritation and redness occur at levels near or below 10-12 mg-min/m³ after a latency period lasting from several hours to several days. Conjunctivitis, tearing, sensitivity to light, and a sensation of grittiness under the eyelids may occur at Cts of 50-100 mg-min/m³ after a latency period of 4-12 hr (Uhde and Dunphy, 1944). Corneal edema and clouding, eyelid edema, photophobia, and severe blepharospasm appear at Cts higher than 100 mg-min/m³, and the ICT₅₀ (the Ct causing incapacitation in 50% of the individuals exposed) has been reported to be 200 mg-min/m³. Exposure to Cts of 400-800 mg-min/m³ are very likely to result in corneal damage and possible ulceration after a latency period of 1-4 hr (see also Geeraets et al., 1977).

Animal Studies. Following severe exposures, the ocular effects seen in laboratory animals are very similar to those occurring in humans and include corneal edema, epithelial necrosis and ulceration, infiltration of polymorphonuclear neutrophils, progressive vascularization and recurrent ulceration after a latency period of years (IOM, 1993). Acute toxicity studies on rabbits indicate that the sulfur mustard-induced ocular effects are caused by the direct contact of the agent with ocular tissue and are not the result of systematic absorption and subsequent transport to the eye (Warthin et al., 1918).

Several studies indicate that rabbits and dogs are less susceptible to the ocular effects of sulfur mustard than humans. Laughlin (1944a) reported that about half the rabbits exposed to a Ct of 200 mg-min/m³ (29-60 min) showed slight conjunctival redness and edema, but no corneal changes, whereas all those exposed to a Ct of 400 mg-min/m³ (18-66 min) exhibited moderate corneal staining and opacity as well as conjunctival redness and edema. Reed (1920) reported that dogs developed a definite conjunctivitis following a 2-hour exposure to a concentration of 1 mg/m³, (Ct of 120 mg-min/m³) but not to a 1-hour exposure to the same concentration (Ct of 60 mg-min/m³). In reviewing an unpublished report from the Medical Research Laboratory at Edgewood Arsenal, Henry (1991) reported that dogs (number of test animals not reported) exposed to 10 mg/m³ for 10 min (Ct = 100 mg-min/m³) exhibited only mild corneal swelling; a 20-min exposure (Ct = 200 mg-min/m³) caused conjunctival and corneal symptoms, and a 40-min exposure (Ct = 400 mg-min/m³) resulted in inflammation of the conjunctiva and lids, edema, opacity and ulceration. Based on comparison with the results of human studies (i.e., conjunctivitis at Cts of 12-70 mg-min/m³), Henry (1991) concluded that the eyes of humans were 3

times more sensitive than the eyes of rabbits and 2 times more sensitive than the eyes of dogs.

Ct^a (mg-min/m³)	Latency Period	Signs and Symptoms	Significance and Duration of Injury
3-10; <12 ^b	Sev. hr to sev. days	Reddening, conjunctivitis	Threshold for signs and symptoms. Nondisabling
20-50 ^c	–	Ocular edema	Tasks requiring intense or prolonged use of eyes may cause rapid fatigue
50-100 ^d	4-12 hr	Conjunctivitis, sensation of grittiness under the eyelids, tearing, sensitivity to light	Healing in 2-7 days, 2 wks in severe cases
200 (ICt ₅₀) ^e	3-12 hr	Corneal edema and clouding, eyelid edema, photophobia, severe blepharospasm leading to temporary blindness	Incapacitating injury. Recovery period several wks
400-800 ^f	1-4 hr	Corneal damage with possible ulceration and secondary infection	Incapacitating injury. Prolonged recovery period of several months. Permanent eye damage in some cases
>800 ^g	1-3 hr	Severe corneal damage, possible permanent loss of vision ^f , possible systemic effects	Incapacitating injury. Prolonged recovery period. Permanent eye damage in some cases

SOURCES: Adapted from Papirmeister et al. 1991, Table 2.4; Barkley, 1999

^a At ambient temperatures of 16-27°C

^b PCS, 1946; McNamara et al., 1975; Sim, 1971; Gates and Moore, 1946; Reed, 1920

^c Reed, 1918; Anderson, 1942

^d McNamara et al., 1975; PCS, 1946; Gates and Moore, 1946

^e PCS, 1946; Urbanetti, 1988; DA/DAF, 1975

^f Karnofsky and Nolen, 1944; exposures were estimated

^g Minimum exposure causing permanent visual impairment in humans is unknown

Exposure of dogs to 0.001 mg/m³ for 24 hr/day, 5 days/wk (method of analysis of vapor concentration not reported) for up to one year produced no eye irritation or injury (McNamara et al., 1975); however, dogs exposed to a time-weighted daily average concentration of 0.029 mg/m³, 5 days/wk for up to one year, exhibited ocular changes (corneal opacity, pannus, chronic keratitis, vascularization, pigmentation, and granulation) 16 wk or more after the exposures were initiated (see Section 2.3.6.2 for a more detailed discussion). When sulfur mustard is administered parenterally to laboratory animals at dose levels that are systemically toxic and lethal, there is little involvement of the eyes (Papirmeister et al., 1991). Therefore, it is likely that the ocular effects observed in the dogs exposed to sulfur mustard vapors in the McNamara et al. study were due to the direct contact of the agent on the corneal/conjunctival epithelium and not due to systemic uptake.

Laughlin (1944a) evaluated the effects of sulfur mustard vapors on the eyes of rabbits and found that for a given Ct, the effect decreased with increasing exposure time; a Ct administered in 2 min produced slightly more severe effects than the same Ct delivered in 30-60 min, and a 7-hr Ct had to be

twice the 30-60 min Ct to produce the same severity of effect. Laughlin (1944a) also reported that the eyes of rabbits became sensitized to sulfur mustard following high Ct exposures (i.e., 400 mg-min/m³).

2.3.4.2 Effects on the Respiratory Tract.

Signs and Symptoms of Exposure. The effects of sulfur mustard on the respiratory tract include irritation of the nasal mucosa, hoarseness, sneezing, burning pain of the mouth as well as nostrils and pharynx, frontal or ethmoid sinus pain, rhinorrhea, epistaxis (nosebleeds), sore throat, tracheobronchitis, tachypnea, dysphonia, and cough (Papirmeister et al., 1991; Somani, 1992; IOM, 1993; Barkley, 1999 and colleagues). Inflammatory reactions leading to epithelial necrosis can result in exudation and the formation of diphtheritic-like pseudomembranes in the trachea and bronchi. Such pseudomembranes may slough off and obstruct the airways. The latency period for the development of respiratory tract effects is usually longer than that associated with ocular effects.

Respiratory infections are often a secondary complication following sulfur mustard-induced injury, and pulmonary edema and bronchopneumonia may develop (Papirmeister et al., 1991; Sidell and Hurst, 1992; IOM, 1993). Although sulfur mustard affects primarily the upper respiratory tract, in severe cases of exposure, such as during the Iran-Iraq conflict, the lower airways and lung parenchyma may also be affected (Hosseini et al., 1989). IOM (1993) reported that there was sufficient evidence to indicate a causal relationship between exposure to "sufficient concentrations" of sulfur mustard and chronic respiratory problems including chronic bronchitis, emphysema, and asthma.

Exposure-Response Data. Exposure-response data, as summarized by Papirmeister et al. (1991), IOM (1993) and Barkley (1999), are presented in Table 4. As shown, the lowest cumulative exposures (Cts) causing a noticeable effect are in the range of 12-70 mg-min/m³. In contrast, mild ocular effects may occur at Cts less than 12 mg-min/m³ (Table 3). The LC_{t50} for inhalation exposures in humans is estimated to be 1000-1500 mg-min/m³. This is not substantially different from the LC_{t50} values reported for laboratory animals (see next subsection).

Animal Data. The respiratory tract lesions seen in animals exposed to acutely toxic vapor concentration of sulfur mustard are similar to those found in humans, with damage occurring to the nasal passages, pharynx, larynx, trachea, bronchi, and in some cases to the bronchioles (IOM, 1993). Unlike the case in humans, however, chronic bronchitis is difficult to induce in laboratory animals following cessation of exposure. In dogs and rabbits, low-level exposures often produce small ulcerations in the trachea and larynx, with subsequent formation of scar tissue leading to contraction of these parts of the upper airway (IOM, 1993). Thus, low level exposures in common species of laboratory animals are most likely to affect higher sections of the respiratory tract than is the case in humans.

The effects of low concentrations of sulfur mustard vapor on the breathing pattern of mice was investigated by Vijayaraghavan (1997) who exposed test animals for 1-hr periods in a head-only exposure chamber to 8.5, 16.9, 21.3, 26.8, 42.3, or 84.7 mg/m³. The animals were monitored for 7 days post-exposure for signs of sensory irritation, airflow limitation, and pulmonary irritation. Sulfur mustard induced sensory irritation, but not pulmonary irritation, and there was a concentration-dependent decrease in respiratory frequency and an increase in tidal volume. The RD₅₀ (concentration causing a 50% decrease in respiration) was estimated to be 27.4 mg/m³, and the LC₅₀ was estimated to be 42.4 mg/m³. The ratio of flow/tidal volume was decreased at 26.8 and 42.5 mg/m³.

Median lethal Ct values for sulfur mustard range from 600 to 1900 mg-min/m³ for 10-min exposures (see Gates and Moore, 1946; Rosenblatt et al., 1975 for reviews). An LC_{Lo} (lowest lethal concentration) of 189 mg/m³/10 min has been reported for mice (Lewis and Sweet, 1984), and a 5-min LC_{Lo} of 77 ppm has been reported for dogs (ITII, 1975).

Table 4. Cts for Respiratory Tract Effects in Humans

Ct ^a (mg-min/m ³)	Latency Period	Signs and Symptoms	Significance and Duration of Injury
12-70 ^b 5-25 ^b	12 hr- 2 days	Irritation of the nasal mucosa, hoarseness	Recovery period may last 2 wk.
200 (IC _{t50}) ^c	4-6 hr	Upper airway: sneezing, lacrimation, rhinorrhea, epistaxis, sore throat, and hoarseness Lower airway: tracheobronchitis, hacking cough, tachypnea. Pulmonary edema and bronchopneumonia may develop after 36-48 hr.	Prolonged recovery (1-2 mo after secondary infections)
1000-1500 (LC _{t50}) ^d	1-4 hr	Injury described as above, progressing to edematous changes in pharynx and tracheobronchial tree; possible death due to secondary bacterial infections, necrotic bronchopneumonia, or airway edema and obstruction	Severe and incapacitating injury for survivors; convalescence of several mo.

SOURCE: Adapted from Papirmeister et al., 1991 (Table 2.5), IOM, 1993 (Table 7-1), and Barkley, 1999.

^a At ambient temperatures of 16-27°C

^b PCS, 1946 reported mild respiratory symptoms in some subjects; Sim, 1971 reported no significant effects at 60 mg-min/m³. Barkley (1999) and colleagues estimated nasal mucosal irritation in range of 5-25 mg-min/m³ and rhinitis at 20-50 mg-min/m³ from data in Reed, 1918; Reed et al, 1918; Reed, 1920 and Anderson, 1942.

^c Ganas, 1969

^d Stepanov and Popov, 1962; DA/DAF, 1975; WHO, 1970; LC_{t50} values presumably estimates based on animal data

2.3.4.3 Effects on the Skin.

Signs and Symptoms. Depending on the concentration and the region of the body affected, exposure of the skin to sulfur mustard vapors can result in erythema, itching, sensitivity to touch, burning sensations, edema, the formation of pinhead-sized vesicles coalescing into blisters, and the development of ulcerous and necrotic lesions after the blisters rupture. Secondary skin infections and systemic toxicity can develop in cases of severe exposures. Factors influencing the severity of effects include ambient temperature and humidity, perspiration, the site and thickness of the skin, and possibly the age and gender of the individual (Papirmeister et al., 1991; Barkley, 1999 and colleagues). Increase in temperature and humidity produce a more severe response presumably by enhancing skin penetration and absorption. Children and women may be more susceptible than men because of their thinner skin (IOM, 1993). It has also been suggested that there may be inherent genetic factors that predispose certain individuals to sulfur mustard-induced skin injury (Papirmeister et al., 1991); however, IOM (1993) has noted that there is no good experimental data indicating that skin color will alter the severity of the response.

Exposure-Response Data. Ct-related skin effects, as summarized by Papirmeister et al. (1991) and Barkley (1999) and colleagues, are presented in Table 5. The threshold for erythema has been estimated to approximate 50 mg-min/m³. This Ct is similar to the threshold for respiratory tract effects (12-70 mg-min/m³), but higher than the reported threshold for ocular effects (<12 mg-min/m³). Cumulative exposures of 100-300 mg-min/m³ result in moderate levels of erythema but no blister formation. The ICT₅₀ (estimated concentration x time exposure profile which is incapacitating to 50% of exposed individuals) is 1000-2000 mg-min/m³. The LCt₅₀ for skin exposures has been estimated to be 10,000 mg-min/m³ (DA, 1974; NDRC, 1946).

Ct^a (mg-min/m³)	Latency Period	Signs and Symptoms	Significance and Duration of Injury
50 ^b , 30-100 ^b	4-12 hr	Mild erythema	Threshold for signs and symptoms.
>100-300 ^c	4-8 hr	Erythema, itching, sensitivity to touch, genital burns and scrotal edema	Healing in 5-20 days
1000-2000 (ICT ₅₀) ^d	3-6 hr	Severe erythema, followed at approximately 12-24 hr by blistering	Incapacitating injury. Recovery period several wks to several mo.
10,000 (LCt ₅₀) ^e	1-3 hr	Rapid development of erythema, followed in 3-12 hr by severe blistering and concomitant systemic intoxication	Incapacitating injury for survivors. Prolonged recovery period

SOURCES: Adapted from Papirmeister et al., 1991 (Table 2.1); Barkley, 1999

^a At ambient temperatures of 16-27°C

^b PCS, 1945, 1946; Reed, 1920

^c PCS, 1946; Stepanov and Popov, 1962

^d PCS, 1946; DA/DAF, 1975

^e DA/DAF, 1975; NDRC, 1946; LCt₅₀ value presumably an estimate based on human and animal data

In the case of exposures to liquid sulfur mustard, doses up to 50 µg/cm² may cause erythema, edema, and sometimes small vesicles. Doses of 50-150 µg/cm² cause bullous-type vesicles, and larger doses cause necrosis and ulceration with peripheral vesication. Ward et al. (1966) reported that droplets of liquid sulfur mustard containing as little as 0.0025 mg may cause erythema. Sidell and Hurst (1992) state that a droplet of 10 µg is sufficient to cause vesication, and, assuming that 80% evaporates and 10% enters the circulation, they conclude that the amount causing the effect may be as little as 1 µg. In summarizing the results of a series of dermal toxicity studies in which 209 men were exposed to droplets of pure sulfur mustard (at temperatures of 64-72 °F and relative humidities of 25-40%), Landahl (1945) reported that a dose of 2.5 µg caused erythema in 87 men and blistering in 5. Renshaw (1946) reported that absorption of 5-20 µg/cm², with 1 or 2 µg/cm² becoming fixed, could cause vesication. The LD_{Lo} for skin exposure is reported to be 64 mg/kg body weight, and the LD₅₀ is estimated to be about 100 mg/kg (DA, 1974, 1991).

Cultured human epithelial cells treated with sulfur mustard display a clear dose response of cell cycle disruption, DNA fragmentation and repair (Emison and Smith, 1997). Assays of primary human

epidermal keratinocytes (HEK) and HeLa cells treated with sulfur mustard at concentrations of 0, 3 μ M or 250 μ M document cell cycle disruption at both dose levels but at different points in the cycle. At a dose of 3 μ M, a quantity less than the sulfur mustard concentration that produces vesicles on human skin (>100 μ M; Smith et al 1990, 1993), cell cycle disruption occurred at the G2/M phase (tetraploid phase following DNA synthesis, but prior to mitosis; Lewin, 1990). However, at 120 hr postexposure, cell-cycle progression in the 3 μ M sulfur mustard-treated cells had returned to normal. At a dose of 250 μ M sulfur mustard, a quantity in excess of that causing vesication on human skin, the cell cycle became blocked at the G1 phase (period preceding DNA synthesis when cell is in diploid phase; Lewin, 1990) and did not return to normal even after 144 hr postexposure. In addition, a large percentage of cellular DNA was fragmented, and cell death occurred in the cells treated with 250 μ M. These results indicate that human epithelial cell recovery occurs after sulfur mustard exposure providing that the exposure is less than that known to induce vesication, and sufficient time is allowed for cell recovery mechanisms to develop.

Animal Data. Median lethal levels in laboratory animals following cutaneous exposures range from 5 to greater than 168 mg/kg body weight (Anslow and Houck, 1946). Factors possibly affecting lethality estimates include the site of application, whether volatilization of the agent was prevented, and whether the animals were prevented from licking the site and ingesting the agent.

2.3.4.4 Sensitization.

Sensitization reactions to sulfur mustard have been reported, primarily following skin exposures; however, as noted by McNamara et al. (1975), “As a general rule, chemical sensitization occurs only after detectable insult” (in context, this statement is interpreted to mean an observable effect, i.e., skin burn, rather than simply an analytically detectable concentration).

Skin. Skin sensitization and hypersensitization reactions to sulfur mustard have been studied in humans and animals (Moore and Rockman, 1950; see also earlier reviews by Sulzberger et al., 1945 and Renshaw, 1946). Sensitization reactions are commonly of the eczematous type (erythematous, papular and vesicular) occurring in 24-48 hr, but they may also be of the urticarial type (wheal forming) within minutes of exposure (Sulzberger et al., 1945). Sensitization may be expressed in terms of a reduced overall tolerance to the agent, or an enhanced response, particularly at a previous burn site. Moore and Rockman (1950) reported that some test individuals who exhibited only an erythematous reaction following an initial skin exposure developed a hypersensitive response at the same site when exposed a week later at a different site. This flare response was seen in about 25% of the men tested, but a general increased sensitivity to the agent was not observed. In reviewing earlier human studies, Sulzberger et al. (1945) note that 30-65% of the test subjects develop some degree of skin hypersensitivity to sulfur mustard when applied as a drop of dilute solution in benzene directly to the skin (Porton Reports, 1931a, 1931b, as cited in Sulzberger et al., 1945). The degree of sensitization observed depended on the magnitude and frequency of previous burns, and was, in one case as much as 1000 times greater than “normal” (Sulzberger et al., 1945). Whether skin sensitization occurs at subsymptomatic exposure levels has not been clearly documented. Papirmeister et al. (1991) cite an anecdotal account (Otto, 1946) indicating that “low dose exposure” to sulfur mustard may cause an increased sensitivity to later exposures; however, neither exposure levels, durations, or conditions were described.

Studies conducted on guinea pigs indicate that sensitization to sulfur mustard can occur following application of the agent directly on the skin (see Sulzberger et al., 1945, and Renshaw, 1946

for reviews). However, long term exposure to low concentrations (too low to cause skin erythema, scaling, crusting or vesication) do not appear to cause skin sensitization in laboratory animals. Guinea pigs who had been vapor-exposed to 0.029 mg/m³ (TWA) for one year and then skin challenged with 7.9 µg of sulfur mustard in castor oil showed no evidence of skin sensitization (McNamara et al., 1975). Subsequent tests of the same animals with 63.2 µg and 31.6 µg (on different sites) elicited the same response as that seen in the controls. Guinea pigs who had been skin-sensitized with liquid sulfur mustard (280 mg dissolved in ligroin 5 days/wk for 3 weeks) did not show an enhanced skin response when exposed to 0.1 mg/m³ for 3 days.

Respiratory tract. Some occupational exposure studies suggest that sulfur mustard exposure may induce respiratory tract sensitization, and Papirmeister et al. (1991) note that hypersensitivity reactions in the respiratory tract are likely considering that hypersensitization does occur in the case of skin exposures. However, in tests on dogs, McNamara et al. (1975) found that exposures to 0.029 mg/m³ (TWA) for up one year had no effect on respiratory rate and volume, suggesting that sensitization had not occurred.

Ocular. There are no experimental human data evaluating the occurrence of ocular sensitization to sulfur mustard. Animal data suggest that ocular sensitization occurs following exposures that produce severe effects. McNamara et al. (1975) cite an earlier study by Laughlin (1944a) in which rabbits were exposed to a sulfur mustard Ct of 400 mg-min/m³. Two weeks later when the eyes appeared normal and the exposure was repeated, the response was more severe. However, in tests in which liquid mustard (0.02-200 µg in ligroin) was applied to the eyes of rabbits, McNamara et al. (1975) found no signs of sensitization two weeks later when a dose of 2 µg was applied. McNamara et al. (1975) reported no signs of increased ocular sensitivity in dog or guinea pigs exposed for 1 yr to 0.029 mg/m³ (TWA), and no generalized hypersensitization reaction, as indicated by the release of bradykinin or histamine in the plasma, was seen in dogs exposed to 0.029 mg/m³ (TWA) for six months.

2.3.4.5 Systemic Effects.

Gastrointestinal, hematological, and neurological effects can occur after acute high exposures to sulfur mustard (IOM, 1993). Exposed individuals may exhibit anorexia, malaise, nausea, vomiting, fever, mental depression, leucopenia, thrombocytopenia, and anemia, (Papirmeister et al., 1991; IOM, 1993). Gastrointestinal effects may be due to inflammatory reactions, delayed radiomimetic effects on the small intestine, or physical stress secondary to skin injury or other effects (Papirmeister et al., 1991). Mustard-induced aplastic or hypoplastic bone marrow can result in immunosuppression and a subsequent increase in the incidence of infectious disease (IOM, 1993). Immunological abnormalities, such as depressed lymphocyte mitogen response (to phytohemagglutinin) and changes in lymphocyte subsets have been observed in former employees of a Japanese chemical agent manufacturing plant who had been exposed to sulfur mustard and/or lewisite (Yamakido et al., 1986a and 1986b). Unlike the case for severe exposures, there is very little information on the potential for mustard-induced systemic effects following long-term exposures to low and subsymptomatic concentrations.

2.3.5 Subchronic Toxicity

Exposures to Sulfur Mustard Vapors. As part of a long-term animal inhalation study, McNamara et al. (1975) exposed five species of animals to two different sulfur mustard vapor

concentrations for time periods varying from 1 to 52 weeks. A full description of the experimental protocol and the results of this study are given in Section 2.3.6.2.

Oral Exposures. In a subchronic study conducted by Sasser et al. (1989a), Sprague-Dawley rats (12/sex/group) were dosed by gavage with 0, 0.003, 0.01, 0.03, 0.1 or 0.3 mg sulfur mustard (in sesame oil)/kg body weight/day, 5 days/week, for 13 weeks. No mustard-related mortality occurred at any dose level. Body weights were significantly decreased in animals in the high-dose group. Epithelial hyperplasia of the forestomach occurred in 5/12 males and 5/12 females of the high-dose group and in 1/12 males receiving 0.1 mg/kg/day, but not in any other treatment group. Forestomach lesions were not seen in any of the control animals. No other treatment-related pathological lesions, clinical chemistry changes, or hematological abnormalities were reported.

2.3.6 Chronic Toxicity

2.3.6.1 Human Data

Limited information on the chronic toxicity of sulfur mustard comes from early studies of workers at research laboratories and at chemical agent manufacturing and munitions plants. Evaluation of these studies is complicated by the fact that, in many cases, the workers were exposed to multiple toxic chemicals and the exposures may have been, at times, sufficiently high to cause acute effects. In addition, the studies of these workers were often lacking in comparisons to control data, were usually not adjusted for confounding factors such as age and smoking history, and usually did not include follow up studies to determine the long-term health effects. Because quantitative exposure data are not available, such studies cannot be used to determine exposure-response relationships and minimum effect levels. Nevertheless, they do reveal that, as in the case of acute exposures, the eyes and respiratory tract are the main target organs affected by chronic exposures to sulfur mustard vapor. Some of these studies are briefly summarized here. The reported carcinogenic effects resulting from occupational exposures are discussed in Section 2.3.10.1.

In a study of 19 workers at a laboratory involved in sulfur mustard research, Laughlin (1944b) found that a high percentage of the individuals exhibited corneal changes, consisting of minute, scattered intraepithelial spots, coalescence of such spots into an amber or brown-colored spindle band in the corneal epithelia; and superficial, white epithelial flecks. In a study conducted on 117 workers at Edgewood Arsenal, Laughlin (1944c) found that 57% of the examined workers exhibited ocular signs. Of these workers, 45% showed a relatively low level of conjunctival "injection", 5% a moderate level of injection, and 4% a high level. In addition, 63% exhibited corneal pigmentation and 18% had corneal staining.

Morgenstern et al. (1947) reported that 3 weeks to 12 months after beginning employment at a sulfur mustard munitions plant some workers developed clinical signs of exposure including "red eyes", photophobia, lacrimation, impaired vision, blepharospasm, loss of taste and smell sensation, nose bleeds, sore throat, difficulty in swallowing, hoarseness, chest pains, retrosternal soreness, wheezing, and dyspnea. In addition, some individuals also exhibited anorexia, vomiting, weight loss, general weakness, insomnia, and irritability. After the affected workers were removed from the source of exposure, most of the clinical signs and symptoms, including the reported ocular problems disappeared; however, a persistent, hacking, productive cough remained and was accompanied by wheezing and chest tightness and, in some cases by dyspnea on exertion. Many of these individuals developed chronic

bronchitis and, in some cases, bronchiectasis (chronic dilation of the bronchi and bronchioles). Morgenstern et al. (1947) state that these case histories show that “prolonged exposure to low concentrations” of sulfur mustard can cause bronchitis leading to partial or total disability. The actual sulfur mustard vapor concentrations to which these individuals were exposed is not reported, nor is it indicated whether the workers were required to wear respiratory protection. The severity of the clinical signs exhibited by the affected workers suggests that episodes of acute toxicity had occurred.

As a follow-up to the report of Morgenstern et al. (1947), Brown (1949) reported that individuals developing chronic pulmonary effects (cough, chest pain, shortness of breath, fatigue, classical bronchiectasis and progressive emphysema) while working at the mustard filling plant all experienced one or more episodes of acute exposure.

In a retrospective study of workers who had been employed at a factory manufacturing sulfur mustard (as well as lewisite, diphenylcyanarsine, hydrocyanic acid, chloroacetophenone, and phosgene), Wada et al. (1962a, b) found that a large number of individuals exhibited productive cough, irregular fever, chronic bronchitis, emphysematous changes, and pleural adhesions. The exposure levels at this plant were estimated to have reached as high as 50-70 mg/m³ (Inada et al., 1978); therefore, these workers are likely to have had multiple exposures at sufficiently high concentrations to cause acute toxic effects. These workers were reported to have elevated risks of respiratory tract cancers (see Section 2.3.10.1)

2.3.6.2 Animal Studies

McNamara et al. (1975) exposed five species of laboratory animals to sulfur mustard vapors for time periods varying from 1 to 52 weeks. The test animals included male and female SDW (Sprague-Dawley-Wistar) rats (70 each), A/J mice (70 each), rabbits (12 initially, 18 total), guinea pigs (30 initially and 42 total), and dogs (6 initially and 10 total). Two series of bioassays were conducted; a toxicity study and a carcinogenicity study. In both studies the animals were exposed to two different concentrations of sulfur mustard; 0.001 mg/m³ for 24 hr/day or to 0.1 mg/m³ for 6.5 hr followed by 0.0025 mg/m³ for 17.5 hr each day. In both cases the exposures were 5 days/week. The latter exposure protocol is equivalent to a time-weighted average concentration of 0.029 mg/m³ for 5 days (this group was referred to as the 0.1 mg/m³ exposure group by McNamara et al., and this same indicator will be used in the current report). One hundred ICR mice were added to the test chambers about 6 months after the tests began (and exposed for 20 weeks), and 100 A/J mice were added to the chambers about 3 months later (and exposed for 9 weeks). Unexposed controls consisted of 10 dogs, 24 rabbits, 32 guinea pigs, 100 rats and 120 A/J mice. The control animals were housed in the “animal colony” in a separate building. McNamara et al. (1975) do not state if the control animals were kept in an exposure chamber. Two and six-month controls are those animals maintained in the separate quarters and not exposed to sulfur mustard for periods of 2 and 6 months before sacrifice.

Exposed animals were sacrificed periodically during the study and, in some cases, new animals were added to the exposure chambers. The test and sacrifice schedule varied slightly for each species. For each exposure concentration 2 dogs were sacrificed after 4, 8, 16, 32 and 52 weeks of exposure; 1-4 rabbits at 1, 2, 4, 12, 16, 32, and 52 weeks; 1-6 guinea pigs at 1, 2, 4, 8, 16, and 32 weeks; 10-30 rats at 1, 2, 4, 8, 12, 24, 36 and 52 weeks; 10-30 A/J mice at 1, 2, 4, 8, 12, 24, 36 (41 for the low exposure group), and 52 weeks; and 10-25 ICR Swiss mice at 12, and 17 weeks in the 20-wk exposure. Hematology and clinical chemistry measurements were made on rabbits and dogs at the time of sacrifice;

however, statistical analyses of the data were not presented. It was reported that there were no changes in the hematology or clinical chemistry parameters in either species, except for a possible increase in serum aspartate amino transferase in dogs exposed for 12-28 weeks to 0.1 mg/m³.

McNamara et al. (1975) reported that no agent-related toxicity appeared in any of the animals exposed to 0.001 mg/m³. In the tabulated data presented by McNamara et al. (1975), tracheitis, chronic murine pneumonia (CMP) and chronic keratitis occurred in these exposed rats (see Table 6).

Table 6. Toxicity of 0.001 mg/m³ Sulfur Mustard in Rats^a			
Number and sex	Exposure duration (months)	Post-exposure duration (days)	Findings^b
5f	0 (2 mo controls)	0	CMP in 4/5
5m	2	0	Tracheitis in 2/5
5f	2	0	Tracheitis in 2/5; CMP in 2/5
5m	3.	0	Tracheitis in 3/5; CMP in 2/5
5f	3	0	Tracheitis in 1/5; CMP in 1/5
5m, 5f	6	0	Chronic tracheitis in 4/10; CMP in 7/10
5m, 5f	0 (6 mo. controls)	0	Chronic tracheitis in 1/10; CMP 8/10
5m	8	0	CMP in 2/5
5f	8	0	CMP in 4/5
5m	12	0	Chronic tracheitis in 1/5 Chronic nephritis in 1/5
5f	12	0	Fibroadenoma in 1/5
4m	12	90	Chronic keratitis in 3/4; hepatic microabscesses in 1/4; CMP in 1/4
5f	12	90	Chronic keratitis in 2/5
14f	12	180	Follicular tracheitis in 5/14; skin papilloma in 1/14; CMP in 2/14
6m	12	180	Chronic tracheitis in 1/6; endocarditis in 1/6; cholangitis in 4/6

SOURCE: McNamara et al., 1975

^a 0.001 mg/m³ for 24 hr/day, 5 days/wk

^b CMP = Chronic murine pneumonia

Of 39 rats exposed to 0.001 mg/m³ for 12 months, 5 exhibited chronic keratitis, a condition marked in the original McNamara et al. (1975) Table A-30 as being possibly agent-related. This effect was also observed in one control rat and in one animal exposed to 0.1 mg/m³ (see Table 7).

Number and sex	Exposure duration (months)	Post-exposure duration (days)	Findings^b
5m	2	0	Tracheitis in 3/5; CMP in 2/5
5f	2	0	Tracheitis in 2/5; CMP in 2/5; pericholangitis in 1/5; pulmonary congestion, edema in 2/5
5m	3	0	No significant lesions
5f	3	0	No significant lesions
5m	3 (controls)	0	Tracheitis in 1/5; CMP in 1/5
5f	6 (controls)	0	Tracheitis in 2/5; CMP in 1/5
5m, 5f	4	0	Tracheitis in 1/10; CMP in 5/10; keratitis in 1/10
5f	8 (controls)	0	CMP in 3/5
5m	8 (controls)	0	CMP in 3/5
5m	8	0	CMP in 3/5; hepatitis in 1/5
5f	8	0	No significant lesions
5m	12	0	No significant lesions
5f	12	0	No significant lesions
5m	12 (controls)	0	Tracheitis in 3/5; CMP in 1/5
5f	12 (controls)	0	CMP in 2/5
4m	12 mo	70	Chronic tracheitis in 4/4; CMP in 4/4; kidney, pyelitis in 1/4; tumors in 4/4
4m	12 mo (controls)	90	Chronic tracheitis in 2/4; CMP in 1/4; nephritis in 1/4; keratitis in 1/4; liver lymphocytic foci in 1/4
4f	12 (controls)	90	Chronic tracheitis in 1/4; CMP in 1/4; kidney, pyelitis in 1/4; chronic nephritis in 1/4; tumors in 1/4
1m	12	90	CMP in 1/1
5f	12	90	Tracheitis in 1/5; CMP in 1/5
4f	12 (controls)	180	Tumors
7m	12 (controls)	180	Chronic tracheitis in 4/7; CMP in 7/7
13f	12	180	CMP in 3/13; tumors
6m	12	180	CMP in 3/6; myocarditis in 2/6

SOURCE: McNamara et al., 1975

m = male

f = female

^a Daily exposure to 0.1 mg/m³ for 6.5 hr followed by 0.0025 mg/m³ for 17.5 hr, 5 days/wk; TWA of 0.029 mg/m³

^b CMP = Chronic murine pneumonia

Keratitis was not reported for any of the control or exposed rats in the carcinogenicity study; therefore, the overall estimated occurrence of keratitis was 1/91 for controls, 5/127 for the 0.001 mg/m³ group and 1/136 for the 0.1 mg/m³ group. The 12-month occurrence of keratitis was 1/29 for controls, 5/39 for the 0.001 mg/m³ group and 0/39 for the 0.1 mg/m³ group. Tracheitis occurred in 7/39 rats exposed for 12 months to 0.001 mg/m³ (control data for the 0.001 mg/m³ group not reported), in 5/39 rats exposed for 12 months to 0.1 mg/m³, and in 13/29 control rats (0.1 mg/m³ test). A clear dose-response was not established for these endpoints. Summary values for the occurrence of tracheitis, keratitis and CMP in rats are given in Table 8.

Table 8. Incidence of Tracheitis, Keratitis, and CMP^a in Rats Exposed to Sulfur Mustard						
Endpoint	Exposure level (Toxicity Study)					
	Control	%	0.001 mg/m ^{3b}	%	0.1 mg/m ^{3c}	%
<u>Tracheitis</u>						
12 mo. exposures only	13/29	34.5	7/39	2.6	5/39	12.8
All exposure periods	14/64	21.9	14/79	17.7	11/79	13.9
<u>Keratitis</u>						
12 mo. exposures only	1/29	3.4	5/39	12.8	0/39	0
All exposure periods	1/64	1.6	5/79	6.3	1/79	1.2
<u>CMP</u>						
12 mo. exposures only	9/29	31.0	3/39	7.7	12/39	30.1
All exposure periods	29/64	45.3	21/79	26.6	26/79	32.9

SOURCE: McNamara et al., 1975, Tables A-30 and A-31 and p. 15

^a Chronic murine pneumonia

^b 0.001 mg/m³ for 24 hr/day, 5 days/wk

^c Daily exposure to 0.1 mg/m³ for 6.5 hr followed by 0.0025 mg/m³ for 17.5 hr, 5 days/wk; TWA of 0.029 mg/m³

As part of the current evaluation, rat keratitis incidence data from the chronic toxicity study (Table 8) as well as the combined dataset (combined incidence from chronic toxicity study and the carcinogenicity study, as summarized above) were subjected to chi-square and Pearson correlation analyses. For both cases, the statistical tests determined that there is no statistical difference in keratitis incidence between exposed and control populations. In addition, there is no correlation of observed rat keratitis incidence with agent exposure for both cases tested.

It was noted by McNamara et al. (1975) that some deaths occurred in the groups exposed to 0.001 mg/m³, particularly in the A/J mice, but the deaths were reported to be related to the "conditions of animal storage" rather than due to the sulfur mustard vapors; there was no correlation between deaths and cumulative Ct.

The only overt signs of toxicity seen in the animals exposed to 0.1 mg/m³ were ocular effects in dogs. These effects consisted of pigmentation and granulation, vascularization, chronic keratitis, corneal

opacity, and pannus, some of which appeared as early as 16 weeks after the initiation of the exposure. No signs of ocular toxicity were seen in any of the dogs exposed to 0.001 mg/m³; however, it should be noted that only 2 animals were exposed for the full 52-week period and only 4 animals were exposed for 32 weeks. The tabulated data presented by McNamara et al. indicate that ocular changes occurred in some animals in the high exposure study as early as 16 weeks after the exposures began (Table 9)

McNamara et al. (1975) concluded that it was "possible" that these effects were agent-related. Pneumonitis occurred in several of the dogs exposed to 0.1 mg/m³ (Table 10), but this condition was also seen in the control animals. Because no other respiratory tract lesions were found, McNamara et al. (1975) suggested that the observed pneumonitis was not agent-related. As shown in Table 9, two dogs exposed to 0.1 mg/m³ for 12 months also exhibited anaphylactic syndrome, gastroenteritis, and petechia. Although these effects were considered by McNamara et al. (1975) to be unrelated to the exposure to sulfur mustard, they are consistent with the known vesicant actions of the agent. It is possible that the sulfur mustard condensed on the fur of the animals and was subsequently ingested as a result of grooming behavior. Gastroenteritis could then have resulted from direct contact of the vesicant with the gastrointestinal epithelium.

2.3.7 Chronic and Delayed/Recurrent Effects from Acute Exposures

There is limited evidence that acute exposures to sulfur mustard may lead to long-term respiratory tract damage manifested as asthma-like conditions, emphysematous bronchitis, and increases in incidence of secondary respiratory infections (bronchopneumonia and tuberculosis) (IOM, 1993). Case and Lea (1955) found that over 80% of a group of 1267 soldiers from the UK who had been exposed to sulfur mustard during World War I also had chronic bronchitis based on medical records evaluated in 1952. In this group, 547 deaths had occurred between Jan. 1, 1930 and December 31, 1952, of which 217 were "classified as due to bronchitis (all types)". It was reported that 21 cases would have been expected in a population of the same size based on the mortality rates for the male population of England and Wales.

Beebe (1960) evaluated the occurrence of respiratory tract disease among a group of U.S. soldiers who had served during World War I. Soldiers who had been exposed to mustard gas exhibited greater mortality from tuberculosis and pneumonia than either of two reference groups, a group of soldiers who had pneumonia and a group of wounded soldiers who had not been exposed to sulfur mustard. Although limited by study design, the results of the study also suggested a much higher incidence of chronic bronchitis in the mustard-exposed group.

Manning et al. (1981) reported a significantly increased incidence of mortality from pneumonia among 428 former workers of a sulfur mustard manufacturing facility. The ratio of observed to expected cases was 2 ($p < 0.05$), based on a comparison with the national mortality rates for England and Wales combined. Deaths due to tuberculosis and bronchitis were also elevated (relative risk 2.1 and 1.3 respectively), but not to statistically significant levels.

Morgenstern et al. (1947) reported that a substantial number of employees of a chemical munitions plant who had symptomatic exposures to sulfur mustard, developed chronic bronchitis and, in some cases, bronchiectasis, leading to partial or total disability. Rates of occurrence of bronchitis and exposure levels were not reported (see Section 2.3.6, for more detailed information).

Table 9. Ocular Effects of Sulfur Mustard in Dogs			
Concentration (mg/m³)	Exposure Period (wk)	No. of dogs affected	Eye effects
0.001 ^a	4	0/10	NE
0.001 ^a	8	0/8	NE
0.001 ^a	16	0/6	NE
0.001 ^a	32	0/4	NE
0.001 ^a	52	0/2	NE
1st Group:			
0.1 ^b	4	0/6	NE
0.1 ^b	8	0/4	NE
0.1 ^b	16	0/2	NE
0.1 ^b	28	2/2	Vascularization and pigmentation
0.1 ^b	40	1/2	Corneal opacity, pannus, chronic keratitis, granulation
0.1 ^b	40	1/2	Vascularization and pigmentation
0.1 ^b	52	2/2	Corneal opacity, pannus, chronic keratitis, granulation
2nd Group:			
0.1 ^b	4	0/4	NE
0.1 ^b	8	0/4	NE
0.1 ^b	16	2/4	Corneal opacity, pannus, chronic keratitis, granulation
0.1 ^b	16	2/4	Vascularization and pigmentation
0.1 ^b	32	2/2	Corneal opacity, pannus, chronic keratitis, granulation
0.1 ^b	52	2/2	Corneal opacity

SOURCE: McNamara et al., 1975, Table A-18, p. 34

NE = No adverse effects

^a 0.001 mg/m³, 24 hr/day, 5 days/wk

^b Daily exposure to 0.1 mg/m³ for 6.5 hr followed by 0.0025 mg/m³ for 17.5 hr, 5 days/wk; TWA of 0.029 mg/m³

Table 10. Toxicity of Sulfur Mustard to Dogs				
No. Animals	Exposure^a (months)	Post-exposure (wk)	Gross findings	Microscopic findings
4	2	5	See microscopic findings.	Splenic infarct, 1/4; Pneumonia, granulomatous, 1/4; Pneumonitis, chronic 1/4
1	4	4	No significant lesions	No significant lesions
1	4 (controls)	4	No significant lesions	No significant lesions
1	4	4	No significant lesions	No significant lesions
1	7.5	4	No significant lesions	Keratitis, pigmentation; Pneumonitis, chronic
1	7.5	4	No significant lesions	Keratitis, chronic
1	7.5 (controls)	4	No significant lesions	Pneumonitis, chronic
1	7.5 (controls)	4	No significant lesions	Pneumonitis, chronic, active
1	12	10	Gastroenteritis; Multiple petechiae; Anaphylactic syndrome	Congestion, liver, spleen, lung; Hemorrhage, pancreas; Ulcerative colitis; Keratitis, chronic; Conjunctivitis, lymphocytic
1	12	10	Anaphylactic syndrome	Gastroenteritis, hemorrhagic; Heart, petechia; Keratitis, acute

SOURCE: McNamara et al., 1975, Table A-37, p. 53

^a Daily exposure to 0.1 mg/m³ for 6.5 hr followed by 0.0025 mg/m³ for 17.5 hr, 5 days/wk (TWA 0.029 mg/m³)

Individuals exposed to doses of sulfur mustard sufficiently high to cause skin lesions often suffer from dermatological abnormalities even after the primary lesions have healed. IOM (1993) reported “the evidence indicates a causal relation between acute, severe exposure to mustard agents and increased skin pigmentation and depigmentation, chronic skin ulceration, scar formation and the development of cancer in human skin”. A 2-year follow-up study on 236 battlefield-exposed individuals from the Iran-Iraq war revealed that 41% suffered from skin abnormalities, and 33% had allergic reactions and pruritis (Balaili, 1986). In 282 patients examined 3 years after wartime exposure 37.6% exhibited hyperpigmentation (Dowlati et al., 1993). Other, very late manifestations were hypopigmentation, xerosis, alopecia, urticaria, nodules, papules, keloids and ulcerations (Dowlati et al., 1993). Thomsen et al. (1998) reported that stinging, burning, itching, and pigmentation changes in the skin, occurred in 50% or more of 10 individuals exposed 8-13 years earlier (five were exposed during

the Iran-Iraq conflict and 5 were Danish fishermen accidentally exposed). Ocular abnormalities (conjunctivitis, photophobia, or impaired vision) were reported in 9 individuals; airway problems (including asthma, bronchitis, infections, dyspnea, cough, dryness, or hoarseness) in seven; and impaired short-term memory in eight. The results were not compared with those of a control population.

Some individuals exposed to sulfur mustard concentrations that are damaging to the eyes are susceptible to delayed recurrent keratitis and corneal ulceration (Papirmeister et al., 1991; IOM, 1993). The condition may reappear 8 to 40 years after recovery from the initial exposure (Dahl et al., 1985), with maximum rates of incidence observed at 15-20 yr (IOM, 1993). Recurrent keratitis has been attributed to damage to the limbal region of the conjunctiva (origin of the corneal stem cells), vascularization of the cornea, deposition of cholesterol, calcium and fat in the cornea, and subsequent movement of these deposits to the surface of the eye where they form secondary ulcerous lesions (IOM, 1993). Significantly, it is the initial damage to the corneal stem cells in the perilimbal conjunctiva that prevents normal regeneration of the corneal epithelium and initiates this process. Whether chronic exposure to sub-symptomatic vapor concentrations of sulfur mustard can result in gradual deficits in stem cell function through inhibition of mitosis, and eventually lead to corneal changes typical of acute exposures is uncertain. However, the results of one chronic study in dogs indicate that corneal changes similar to those caused by acute exposures can appear following 16 weeks of exposure, and without prior visible signs of ocular effects such as conjunctivitis (see Section 2.3.6.2)

2.3.8 Developmental and Reproductive Effects

2.3.8.1 Human Data

Azizi et al. (1995) investigated changes in serum concentrations of reproductive hormones and sperm counts in men who had been exposed to sulfur mustard during wartime. In 16 individuals, serum free and total testosterone and dehydroepiandrosterone were markedly decreased in the first five weeks after exposure; but levels returned to normal by 12 weeks. In 28 of 42 men evaluated one to three years after exposure, sperm counts were less than 30 million cells/mL and follicle-stimulating hormone was increased compared with controls having sperm counts above 60 million cells/mL. Testicular biopsy of the test subjects revealed partial or complete arrest of spermatogenesis.

Taher (1992) reported that 30 of 79 cases of cleft lip and palate among 21,138 newborns at an Iranian hospital during the years 1983-1988 were associated with parental exposure to sulfur mustard during the Iran/Iraq war. IOM (1993) noted that lack of appropriate exposure and epidemiological data, made it unclear whether the reported incidence of these defects was truly elevated relative to other regions in Iran or other parts of the world.

2.3.8.2 Animal Studies

Vapor/Inhalation Exposures. McNamara et al. (1975) reported no increased fetal mortality rate when groups of 10 rat dams were exposed to 0.001 mg/m³, 24 hr/day, 5 days/wk or daily to 0.1 mg/m³ for 6.5 hours followed by 0.0025 mg/m³ for 17.5 hours, 5 days/wk during the first, second, or third week, or for the entire period of gestation. In another study, groups of 10 unexposed female rats were bred to male rats which had been exposed to the same sulfur mustard concentrations for 1, 2, 4, 8, 24,

36, or 52 weeks to gain information on dominant lethal mutagenesis. There was no evidence of mutagenesis, and fetal mortality was considered within normal limits. In neither study was the occurrence of fetal abnormalities reported.

Oral Exposures. In a study conducted by Hackett et al. (1987), sulfur mustard (dissolved in sesame oil) was administered by intragastric intubation to rats and rabbits on gestation days 6-15 (rats) or 6-19 (rabbits). Female rats were dosed with 0, 0.2, 0.4, 0.8, 1.6, 2.0 or 2.5 mg/kg/day in a range-finding study (3-9 animals per dose group of which 2-7 per dose group were pregnant) and with 0, 0.5, 1.0, or 2.0 mg/kg/day in a teratology study (25-27 animals per dose group of which 20-26 per dose group were pregnant). Maternal toxicity and fetal effects are listed in Table 11.

In the range-finding study significant ($p < 0.05$) maternal effects included mortality (1/3) at the highest dose; severe gastric lesions (petechial hemorrhage and sloughing of gastric mucosa) at 2.0 and 2.5 mg/kg/day; and inflamed mesenteric lymph nodes at doses of 0.4 mg/kg/day and higher. Significant decreases in body weight and decreased extragestational weight occurred at 1.6 mg/kg/day and decreased hematocrit at 0.8 mg/kg/day. There were no adverse effects on fetal weight and no evidence of morphological abnormalities in the fetuses. In the rat teratology study, maternal toxicity was evidenced by gastric inflammation at 2.0 mg/kg/day, and inflamed mesenteric lymph nodes at doses of 0.5 mg/kg/day and higher. Decreased body weight and decreased extragestational weight occurred at 0.5 mg/kg/day; decreased hematocrit at 1.0 mg/kg/day; and decreased weight of the placenta and gravid uteri at 2.0 mg/kg/day. Fetal effects included decreased weight in females and hydroureter at 0.5 mg/kg/day; decreased weight of males at 1.0 mg/kg/day; increased incidences of supernumerary ribs, misaligned sternbrae, and reduced ossification of sternbrae at 2.0 mg/kg/day. The investigators reported that the study did not reveal any evidence for a sulfur mustard-induced teratogenic effect in rats because all of the observed fetal changes occurred at dose levels that also produced maternal toxicity. However, fetal effects did occur at the lowest tested dose of 0.5 mg/kg/day and because a NOAEL was not identified, it is possible that fetal effects might occur at a lower dose level in the absence of maternal toxicity.

In the second part of the Hackett et al. (1987) study, rabbits were dosed with 0, 0.5, 1.0, 2.0, and 2.5 mg/kg/day in a range-finding study, and with 0, 0.4, 0.6, or 0.8 mg/kg/day in the teratology study. The number of animals tested was 7-8 per dose group, less than the 12 per dose group recommended by EPA (USEPA, 1984). Dose levels of 0.8 mg/kg/day or higher were lethal to the dams. Damage to the gastric mucosa and enlarged Peyer's patches were observed in animals that received the lowest dose (0.4 mg/kg/day). Depressed body weight, depressed extragestational weight gain, and depressed hematocrit values occurred at 0.8 mg/kg/day. In the range-finding study a significant depression in fetal body weights occurred at a dose level of 2.0 mg/kg/day; however, in the teratology study no significant effects were observed on intrauterine survival, placental and fetal body weights, or incidence of fetal abnormalities. The investigators concluded that the study provided no evidence that sulfur mustard induced a teratogenic effect in rabbits. The NOAELs for maternal and fetal toxicity were reported to be < 0.4 mg/kg/day and > 0.8 mg/kg/day, respectively.

In a two-generation reproductive toxicity study conducted by Sasser et al. (1989b), groups of Sprague-Dawley rats (27 females and 20 males/group/generation) were gavaged with 0, 0.03, 0.1 or 0.4 mg/kg/day. The animals were treated according to the following exposure protocol: male and female rats were dosed 5 times/week for 13 weeks prior to mating and during a 2-week mating period; female rats were dosed daily throughout the 21-day gestation and parturition period; and females were dosed 4-5 times/week during the 21-day lactation period. Males who had mated with females were sacrificed at

Effects		Rat studies		Rabbit studies	
		Range-finding (mg/kg/day)	Teratology (mg/kg/day)	Range-finding (mg/kg/day)	Teratology (mg/kg/day)
Maternal Effects:	mortality	2.5	-	1.0	0.8
	gross lesions:				
	major ^a	2.0	0.5	1.0	0.4
	minor ^b	0.4		0.5	0.4
	decreased weight:				
	body	1.6	0.5	2.0	0.8
extragestational	1.6	0.5	-	-	
extragestational gain	0.4	0.5	-	0.8 ^c	
gravid uterus	-	2.0	-	-	
decreased hematocrit	0.8	1.0	-	0.8	
resorptions	0.4 ^d	-	-	-	
Fetal Effects:	decreased weights:				
	female fetuses	-	0.5	2.0	-
	male fetuses	-	1.0	2.0	-
	placenta	-	2.0	-	-
	fetal morphology				
	misaligned sternebrae	-	2.0 ^e	-	-
	supernumerary ribs	-	2.0 ^e	-	-
	reduced ossification				
	vertebrae	-	0.5 ^{d,e}	-	-
	sternebrae	-	2.0 ^e	-	-
hydroureter	-	0.5 ^{d,e}	-	-	

SOURCE: Hackett et al., 1987

^a Gastric lesions or infections

^b Inflamed mesenteric lymph nodes in rats; enlarged Peyer's patch in rabbits

^c Significantly different from lowest dose group, but not from controls

^d Not significant in the highest dose group

^e Significance based on fetal unit

^f Significance based on litter unit

the birth of their pups; dams who had given birth were sacrificed when the pups were weaned. Male and female F₁ pups received sulfur mustard until they were mated, the females became pregnant, and gave birth. At this point, F₁ males were sacrificed and F₁ dams continued on the dosage schedule until weaning, at which point the study was terminated. Thus, two generations of rats received subchronic exposure to sulfur mustard, with each generation going through a mating cycle. Similarly, two generations of pups were born to parents who had received sulfur mustard. Body weight gain was significantly ($p < 0.05$) lower than control values in the F₁ rats of both sexes born to parents who had received the highest dose of sulfur mustard. There were no significant adverse effects on reproductive parameters at any dose level. However, dose-related lesions of the squamous epithelium of the forestomach (acanthosis and hyperplasia) occurred in both sexes of each treatment group. The lesions were described as mild at the lowest dose level, 0.03 mg/kg, compared with the higher dose groups. The incidence and severity of acanthosis was 0/94 in the controls, 71/94 in the low-dose group, 89/94 in the

mid-dose group, and 94/94 in the high-dose group. Benign neoplasms of the forestomach occurred in 8/94 animals in the 0.1 mg/kg group and in 10/94 animals of the 0.4 mg/kg group. The results of this study indicate that lowest dose tested (0.03 mg/kg/day) is a LOAEL for maternal toxicity.

2.3.9 Genotoxicity

IARC (1975), Fox and Scott (1980) and ATSDR (1992), and Papirmeister et al. (1991) have summarized the available evidence concerning the genotoxicity of sulfur mustard. Because sulfur mustard is a strong DNA alkylating agent, genotoxic effects occur through cross-link formation, inhibition of DNA synthesis and repair, point mutations due to replication or repair errors, chromosome breaks, and chromatid aberrations. Some of these conditions have been observed in humans following exposure to sulfur mustard, others have occurred in various test systems including bacteria, yeast, insects, and mammalian cell cultures.

Human Data. Retrospective studies have been conducted on Japanese workers who had been employed at a chemical agent manufacturing plant from 1929 to 1945. Although sulfur mustard was the main product of the facility, lewisite, diphenylarsine, hydrocyanic acid, phosgene, and chloroacetophenone were also produced there (Inada et al., 1978), and it is not known to what degree these other chemicals contributed to the observed effects. In one study of these workers, Yanagida et al. (1988) found that the frequency of mutations to hypoxanthine-guanine-phosphoribosyl-transferase (HGPRT) deficiency in 28 exposed individuals was significantly elevated when compared with two control groups matched for age and smoking status. One control group consisted of healthy men and the other of individuals with bronchitis. The data also showed that the mutations were significantly more frequent in those workers who had longer exposures. A chromosome study of 16 former workers of this same factory indicated a significantly higher incidence of sister chromatid exchanges (SCE) in peripheral lymphocytes when compared with a control group ($p < 0.03$) (Shakil et al., 1993). Two individuals with chronic myelocytic leukemia had an almost three-fold higher SCE rate than controls and also a high (12.1%) incidence of chromosome abnormalities (Shakil et al., 1993). In an evaluation of the p53 mutations found in lung tumors of these workers, Takeshima et al. (1994) found that the mutations were similar to those in lung tumors of tobacco smokers (the factory workers were also tobacco smokers), however, the prominence of G:C to A:T transitions and the occurrence of double mutations in two of twelve cases suggested that exposures in the chemical agent manufacturing plant did contribute to the development of the lung cancers.

Yamakido et al. (1985) conducted an electrophoretic analysis of blood proteins in children of the former workers of this same Japanese chemical agent manufacturing plant to determine the potential genetic effects of exposure to mustard. In consultation with S. Leffingwell of the CDC, IOM (1993) evaluated these data and determined that the sample size was not large enough to detect induced mutations in these children.

Wulf et al. (1985) reported significant ($p < 0.001$) increases in sister chromatid exchanges in lymphocytes of eleven fisherman who inadvertently netted aging and leaking mustard munitions previously dumped at sea following World War II. Contact with liquid sulfur mustard remaining on the nets caused injuries (skin burns, etc.) to exposed fisherman. Although there are no quantitative dose data from Wulf et al (1985), these exposures are considered high due to the severity of sustained injuries.

Other Studies. Sulfur mustard has been found to be genotoxic and mutagenic in several microbial assays. The agent caused alkylation of DNA in the yeast *Saccharomyces cerevisiae* (Kircher and Brendel, 1983), and interstrand DNA cross-links (Venitt, 1968) and inhibition of DNA synthesis (Lawley and Brookes, 1965) in *Escherichia coli*. Using the histidine reversion assay, Stewart et al. (1989) found that sulfur mustard induced point mutations in *Salmonella typhimurium* strain TA102 and frameshift mutations in TA 97, but neither type of mutation in strains TA98 and TA100.

Sulfur mustard inhibited DNA synthesis in mouse lymphoma cells (Crathorn and Roberts, 1965), HeLa cells (Crathorn and Roberts, 1966), and L-strain mouse fibroblasts (Walker and Thatcher, 1968). It also induced chromosomal aberrations in cultured rat lymphosarcoma and mouse lymphoma cells (Scott et al., 1974), and chromosomal aberrations and reverse mutations in male BDF₁ mice in a host-mediated assay using murine leukemia L5178Y/Asn⁻ cell line as an indicator (Capizzi et al., 1973).

Cultured human epithelial cells treated with sulfur mustard display a clear dose response of cell cycle disruption, DNA fragmentation and repair (Emison and Smith, 1997). Assays of primary human epidermal keratinocytes (HEK) and HeLa cells treated with sulfur mustard at concentrations of 0, 3 μM or 250 μM document cell cycle disruption at both dose levels but at different points in the cycle. At a dose of 3 μM sulfur mustard, a quantity less than the sulfur mustard concentration that produces vesicles on human skin (>100 μM sulfur mustard; Smith et al 1990, 1993), cell cycle disruption occurred at the G2/M phase (tetraploid phase following DNA synthesis, but prior to mitosis; Lewin 1990). However, at 120 hr postexposure, cell-cycle progression in the 3 μM sulfur mustard-treated cells had returned to normal. At a dose of 250 μM sulfur mustard, a quantity in excess of that causing vesication on human skin, the cell cycle became blocked at the G1 phase (period preceding DNA synthesis when cell is in diploid phase; Lewin 1990) and did not return to normal even after 144 hr postexposure. In addition, a large percentage of cellular DNA was fragmented, and cell death occurred in the cells treated with 250 μM sulfur mustard. These results indicate that human epithelial cell recovery occurs after sulfur mustard exposure providing that the exposure is less than that known to indicate vesication, and sufficient time is allowed for cell recovery mechanisms to develop.

Several studies have also demonstrated that sulfur mustard causes dominant lethal mutations. Rozmiarek et al. (1973) reported a dominant lethal mutation rate of 9.4% (± ~1.9%) in rats after adult males had been exposed to 0.1 mg/m³ for 12 weeks. Sasser et al. (1990) reported that a dominant lethal effect occurred after male Sprague-Dawley rats were dosed orally with 0.5 mg/kg/day 5 days/week for 10 weeks. The observed effects included increases in early fetal absorptions, preimplantation losses, and decreases in total live embryo implants. A significant increase in the percentage of abnormal sperm was also reported. Dominant lethal mutations, as well as chromosome rearrangements, have also been observed in *Drosophila melanogaster* exposed to sulfur mustard (Auerbach and Robson, 1946).

The cytotoxic, clastogenic and mutagenic effects of sulfur mustard in Chinese hamster ovary cells have been evaluated by Jostes et al. (1989). Chromosomal aberration frequency increased in a dose-dependent manner over the dose range of 0.0625 to 0.25 μM. Mutation induction at the HGPRT locus was sporadic, but the majority of the exposures resulted in mutation frequencies that were 1.2 to 4.0 fold higher than the spontaneous frequencies.

2.3.10 Carcinogenicity

The following section presents studies on workers occupationally exposed to chemical warfare agents, including sulfur mustard; these studies have revealed elevated risks of respiratory tract and skin

tumors after long-term exposure. Data presented in Section 2.3.9 on genotoxicity, and in Section 2.3.10.2 on animal carcinogenicity, along with the fact that sulfur mustard is a potent DNA alkylating agent (IARC, 1987a), provide supporting evidence for the carcinogenicity of this chemical warfare agent in humans.

The International Agency for Research on Cancer (IARC) has classified sulfur mustard as a Group 1 compound (carcinogenic to humans) (IARC, 1987b), and the National Toxicological Program (NTP) first categorized "mustard gas" as a substance "known to be a human carcinogen" in its *First Annual Report on Carcinogens, 1980*. Mustard gas is still listed in the same category in the *Eighth Report On Carcinogens (1998)* (<http://ntp-server.niehs.nih.gov/NewHomeRoc/CurrentLists.html>). The State of Maryland also considers "mustard gas" as a "known human carcinogen" (a Class I.A. Toxic Air Pollutant as defined by the Code of Maryland Regulations, CMR Title 26 Subtitle 11, as amended).

2.3.10.1 Human Data

IARC (1975), Waters et al. (1983), Watson et al. (1989), and the IOM (1993) summarized the epidemiological evidence concerning the potential carcinogenicity of sulfur mustard in humans. Much of this information has come from studies of soldiers exposed during World War I as well as from studies of workers at chemical warfare agent manufacturing facilities. As noted by Papirmeister et al. (1991), the results of retrospective studies are often difficult to interpret because of potential sampling errors; inadequate controls for confounding factors such as smoking, lifestyle, race, sex, age, or exposure to other chemicals; differential quality of available health care; and incorrect diagnosis.

Case and Lea (1955) reported 29 deaths from cancer of the lungs and pleura among a sample of 1267 World War I veterans who had been exposed to sulfur mustard, 80% of whom also suffered from chronic bronchitis. In comparison, 14 cases would have been expected in a population of that size based on the mortality rates for the male population of England and Wales. The mortality ratio (2.07) indicated a highly significantly elevated risk for respiratory tract neoplasms ($p < 0.01$). A similar tumor incidence rate and mortality ratio (2.01) were found in a population of veterans who had never been exposed to mustard but who were suffering from bronchitis. Case and Lea (1955) concluded that the evidence did not support the view that sulfur mustard was a direct carcinogen. It should be noted, however, that studies based on standardized mortality ratios do not address competing risks of death that may be relevant for certain populations. For example, IARC (1975), noted that the high tumor rate in the group not exposed to mustard may have been due, in part, to smoking habits (a significantly higher proportion of men injured by mustard gas had given up smoking by the age of 40).

Beebe (1960) evaluated the occurrence of respiratory tract cancers among a group of 2718 American soldiers exposed to sulfur mustard during World War I and found that the ratio of observed to expected cases was 1.47 (based on U.S. mortality rates) compared with 1.15 for wounded soldiers not exposed to sulfur mustard, and 0.81 for soldiers who had pneumonia, but who had not been exposed to mustard. Norman (1975) evaluated the same group of soldiers after a 10-year follow-up period (study completed in 1965) and found that the exposed men had a 40% excess of lung cancer mortality, with an estimated relative risk of 1.3 (95% confidence limits of 0.9-1.9) compared with a control group consisting of wounded soldiers without exposure to mustard. The latency period was estimated to be 22-37 years. Norman (1975) also reported that in a limited subgroup of veterans, the relative risk of lung cancer mortality among cigarette smokers who were exposed to mustard agents was approximately equal to that of veterans exposed to mustard who stated that they did not smoke (4.3 vs 4.4). Norman (1975)

concluded that there was no evidence in this limited data set that mustard exposure and cigarette smoking had a synergistic effect on lung cancer mortality.

Retrospective studies of Japanese workers who had been employed at a chemical warfare agent manufacturing plant from 1929 to 1945 have revealed that these individuals have an increased risk of developing respiratory tract cancers (see Yamakido et al., 1996, for most recent review). Although sulfur mustard was the main product of the facility, lewisite, diphenylarsine, hydrocyanic acid, phosgene, and chloroacetophenone were also produced (Inada et al., 1978). The concentration of mustard in the workplace was estimated to be as high as 50-70 mg/m³ (Nakamura, 1956), and the workers frequently exhibited signs of mustard toxicity including acute conjunctivitis, acute rhinitis, acute bronchitis, and acute dermatitis with blister formation. Studies completed in the 1950's documented individual cases of bronchial and laryngeal carcinoma in this population of workers (Yamada et al., 1953, 1957). Yamada (1963) reported that 16.3% of 172 deaths of former workers were due to cancers of the respiratory tract and oropharynx. The incidence rate among 5030 non-exposed inhabitants from the same geographic area was reported to be of 0.4% (Yamada, 1963). Mortality rates among the former factory workers during the years 1952-1967 were studied by Wada et al. (1968) who found that the mortality rate due to respiratory tract cancer was 33 individuals out of 495 (30 confirmed by histological evaluation) compared with 0.9 expected deaths, based on national mortality rates for males with the same age distribution as the mustard workers. Of 930 former factory workers not directly involved in the mustard production process, 3 died of respiratory tract cancer compared to 1.8 expected. Neoplasms occurred in the tongue, pharynx, sphenoidal sinus, larynx, trachea, and bronchi; only one occurred peripherally in the lung. The median length of employment at the chemical warfare agent manufacturing facility was 7.4 years, and the median interval between first employment and death from cancer of the respiratory tract was 24.4 years (Wada et al., 1968).

Additional studies of this population of workers were conducted by Nishimoto et al. (1988) who incorporated histopathological and mortality data gathered between 1952 and 1986. For 1632 of these workers, the overall standardized mortality ratio (SMR) for respiratory tract tumors was 3.9 (70 observed vs. 17.8 expected, $p < 0.001$, based on data for the Japanese male population) and the overall SMR for all malignant tumors was 1.2 (173 observed vs. 142 expected, $p < 0.01$). SMRs were also calculated for each of six age groups (Table 12). Age-adjusted SMRs for total malignancies, respiratory tract tumors, and gastrointestinal tract tumors showed significantly higher SMRs for the age-groups from 40 to 80 years. For individuals 30-39 years old the SMRs for respiratory tract cancer were not significantly elevated; however, the SMRs for the 40-49, 50-59, 60-69, and 70-79 yr olds were 10.3, 3.9, 4.4, and 2.5, respectively; all statistically significant at $p < 0.01$ or $p < 0.001$. Adjustments for smoking and post-employment history of exposure to other chemicals were not reported.

The same cohorts were divided into three subgroups; (A) those directly involved in the manufacture of sulfur mustard or lewisite; (B) those not involved in mustard or lewisite manufacture, but who experienced some exposure; and (C) those engaged in the manufacture of other agents and those who were never exposed. The SMR for groups A and B (1.6 and 1.9) were also significantly elevated ($p < 0.001$) whereas that for group C was not. Nishimoto et al. (1988) also found that the SMR was about 2.7 for individuals who had worked at the factory 0.5 to 5 years, but 7.17 for individuals who had been employed for more than 5 years. The SMR was not significantly elevated for individuals who had worked at the factory for 7 months or less.

Tumor types	Category	Age Group (yrs)					
		30-49	40-49	50-59	60-69	70-79	80
Total	Observed	0	11	39	63	49	11
	Expected	1.59	9.89	34.25	54.52	36.79	6.58
	SMR ^a	0	1.1	1.1	1.2	1.3*	1.7
Respiratory tract	Observed	0	8	15	31	15	1
	Expected	0.31	0.78	3.83	7.07	5.94	0.89
	SMR	0	10.3***	3.9***	4.4***	2.5**	1.1
G.I. tract	Observed	0	2	15	18	21	3
	Expected	0.75	5.17	16.14	24.55	15.36	2.57
	SMR	0	0.4*	0.9	0.7	1.4	1.2

SOURCE: Nishimoto et al., 1988

^a Standardized Mortality Ratio; number observed/number expected: * p <0.05, ** p<0.01, *** p <0.001

Data on this same group of workers followed up to 1992 has been summarized by Yamakido et al. (1996). The results do not differ substantially from those of Nishimoto et al. (1983, 1988). The SMRs for total malignant neoplasms and for lung cancer were significantly elevated for Group A and B workers employed in the factories for 5 or more years and also for Group A workers who had been employed for 0.5-5 yr, but not for Group C workers, regardless of their length of employment. The expected number of cases of cancer were derived from Japanese national mortality data, and age-adjusted SMRs were not reported. The SMRs are listed in Table 13.

Histopathological studies conducted by Yamada (1974, as reported by Inada et al., 1978) on 94 autopsy cases and 8 surgical cases revealed 17 cases of gastrointestinal tract cancers among these workers (no comparisons with control groups were reported). Recently Yamakido et al. (1996) reported 85 cases of gastrointestinal tract neoplasms in Group A (12.6% of the 674 survivors in 1952); 62 in Group B (10.4% of the 598 survivors in 1952); and 37 in Group C (10.3% of the 360 survivors in 1952). The total incidence of gastrointestinal tract neoplasms amongst all 1632 survivors from 1952 was 11.3%. The SMR for gastrointestinal tract cancer for all the factory workers was not, however, significantly different from the national average (Yamakido et al., 1996).

Of 488 former workers who were examined dermatologically, 115 had abnormal pigmentation and 22 had skin tumors of which 8 were cases of Bowen's disease (intra-epidermal squamous cell carcinoma) (Inada et al., 1978). Pigmentation disorders were present in 57 cases out of 109 engaged only in the production of mustard and in only 1 of 16 cases engaged only in the production of lewisite. Hyperkeratotic skin lesions such as Bowen's disease, basal cell carcinomas, and hyperkeratotic papular eruptions, were present in 14 cases out of 109 engaged only in mustard production and in 1 case out of 16 engaged only in lewisite production. No abnormalities were observed in 77 former factory workers who had no exposure to chemical agents (Inada et al., 1978). It was also observed that the longer an individual had been exposed to mustard, the more marked the skin lesions tended to become.

Table 13. SMRs^a for Japanese Chemical Warfare Agent Factory Workers: 1952-1992			
Group	Duration of Work		
	#0.5 yr	0.5-5.0 yr	\$5 yr
Total Malignant Neoplasms:			
A	0.78	1.44*	2.36**
B	1.56	1.23	1.66*
C	0.36	0.9	0.75
Lung cancer:			
A	2.32	3.24**	7.35**
B	3.84	2.53	4.92**
C	0	1.08	1.5

SOURCE: Yamakido et al. (1996)

^a SMR: Standardized Mortality Ratio, number of observed cases/number of expected cases from all Japanese mortality data; *p <0.05; **p<0.001.

The studies of Yamakido et al. (1996), Nishimoto et al. (1988), Yamada (1974) and Inada et al., (1978) provide strong evidence for a causal link between chemical agent exposure and cancer of the respiratory tract; however, because the workers were potentially exposed to lewisite as well, it is not possible to state conclusively that the cancers were due solely to sulfur mustard. Furthermore, it should be noted that several possible confounding factors, such as tobacco smoking habits, pre-existing health conditions, and post-exposure occupational histories of the workers, were not evaluated. In addition, the SMR may not provide a good estimate of cancer risk, because it does not take into account the impact of medical intervention and social/economic factors that can affect survival rates.

Weiss and Weiss (1975) conducted studies evaluating the health of 271 workers employed for varying lengths of time between 1935-1945 at a munitions depot where the production, testing and destruction of sulfur and nitrogen mustard (as well as bromoacetone, phosgene, chloropicrin and organic arsenicals) had occurred. Ninety percent of the group had chronic health problems and 114 had died by the end of 1974. Thirty-five percent died from cancer of which 38% were bronchial cancers. The total number of deaths from cancer was significant (p<0.01) and the number of bronchial cancers was also significant (11 observed vs. 5 expected for the population of the geographic region where the facility was located). The number of cancers of the gastrointestinal tract was 35% greater than expected. The average tumor induction time was 21.6 years. IARC (1975) noted that the study was limited to workers with available medical records, which "raises the possibility that the proportion with cancer may have been inflated, since medical records or autopsy records would more likely have been preserved for workers with cancer". Furthermore, IARC (1975) does not mention whether Weiss and Weiss (1975) accounted for smoking habits and other confounding factors.

According to Klehr (1984), German workers involved in the dismantling of a sulfur mustard facility developed multiple skin lesions including basal cell carcinomas, Bowen's disease, Bowen's

carcinomas, and carcinoma spinocellulare. The incidence rate for all tumors (including skin tumors) was 34% in 53 workers evaluated.

Manning et al. (1981) evaluated the incidence of cancer among former workers of a British mustard manufacturing facility (1939-1945). As of 1974, the number of deaths from all neoplasms combined (45) was slightly greater than that expected from national death rates, but the increase was not statistically significant. Two deaths were attributed to cancer of the larynx and one to carcinoma of the trachea, compared with an expected number of 0.40 ($p < 0.02$; relative risk 7.5). Seven individuals were known to have developed cancer of the larynx, compared with 0.75 expected ($p < 0.001$; relative risk 9.3). Lung cancer deaths were also elevated (21 observed vs. 13.43 expected) but not to significant levels (relative risk 1.6). In follow-up investigations of this cohort, Easton et al. (1988) evaluated the mortality records of 3354 workers and found greater numbers of cancer deaths when compared to national mortality rates. Significant increases were observed in deaths from cancer of the larynx (11 observed, 4.04 expected, $p = 0.003$), pharynx (15 observed, 2.73 expected, $p < 0.001$), and all other buccal cavity and upper respiratory sites combined (12 observed, 4.29 expected, $p = 0.002$). There were also 200 deaths from lung cancer compared with 138.39 expected ($p < 0.001$). It was also reported that the risks of developing cancer of the lung and pharynx were significantly related to the duration of employment. Significant excess mortality was also observed for cancers of the esophagus (20 observed vs. 10.72 expected) and stomach (70 observed vs. 49.57 expected) but there was no correlation with time since first exposure or duration of exposure.

Manning et al. (1981) concluded that it was very likely that the observed cancers of the pharynx, larynx and other upper respiratory sites were due to exposure to sulfur mustard because the excesses were too large to be accounted for by confounding factors (the effects of smoking, however, were not evaluated), increased with increasing duration of employment, and were limited to the period more than 10 years after first employment. Evidence for a causal relationship between sulfur mustard exposure and other cancers, including lung cancer, was not considered to be as strong.

Although a large number of American military personnel were exposed to sulfur mustard in chamber and field tests conducted during World War II, the morbidity and mortality records of this cohort have not been adequately evaluated to document long-term health risks (IOM, 1993).

2.3.10.2 Animal Studies

Information on the potential carcinogenicity of sulfur mustard in laboratory animals is available primarily from studies on rats and mice.

In a subchronic study conducted by Sasser et al. (1989a), Sprague-Dawley rats (12/sex/group) were dosed by gavage with 0, 0.003, 0.01, 0.03, 0.1 or 0.3 mg sulfur mustard (in sesame oil)/kg body weight/day, 5 days/week, for 13 weeks. Epithelial hyperplasia of the forestomach occurred in 5/12 males and 5/12 females of the high-dose group and in 1/12 males receiving 0.1 mg/kg/day, but not in any other treatment group. Forestomach lesions were not seen in any of the control animals.

In a two-generation reproductive toxicity study conducted by Sasser et al. (1989b), groups of Sprague-Dawley rats (27 females and 20 males/group/generation) were gavaged with 0, 0.03, 0.1 or 0.4 mg/kg/day. The animals were treated according to the following exposure protocol: male and female rats were dosed 5 times/week for 13 weeks prior to mating and during a 2-week mating period; female

rats were dosed daily throughout the 21-day gestation and parturition period; and females were dosed 4-5 times/week during the 21-day lactation period. Dose-related lesions of the squamous epithelium of the forestomach (acanthosis and hyperplasia) occurred in both sexes of each treatment group. The lesions were described as mild at the lowest dose level, 0.03 mg/kg, compared with the higher dose groups. The incidence and severity of acanthosis was 0/94 in the controls, 71/94 in the low-dose group, 89/94 in the mid-dose group, and 94/94 in the high-dose group. Benign neoplasms of the forestomach occurred in 8/94 animals in the 0.1 mg/kg group and in 10/94 animals of the 0.4 mg/kg group. The results of this study indicate that lowest dose tested (0.03 mg/kg/day) is a LOAEL for maternal toxicity.

Heston (1950) reported an increase in the occurrence of pulmonary tumors in strain A mice injected intravenously with 0.25 mL of a 1:10 dilution of a saturated solution of sulfur mustard in water (0.06-0.07%) at 2-day intervals for a total of 4 doses. The tumor incidence was 93.3% with 2.6 tumors/mouse compared with 61% and 0.9 tumors/mouse in the controls. In a second test in which a slightly lower dose was used, pulmonary tumors were found in 68% of the surviving treated animals (1.09 tumors/mouse) compared with 13% in the controls (0.13 tumors/mouse) ($p < 0.001$). A significant increase in the incidence of pulmonary tumors in strain A mice was also seen in an inhalation study in which the test animals were exposed for 15 min to vapors released from 0.01 mL of sulfur mustard applied to filter paper (Heston and Levillain, 1953; exposure levels were not otherwise quantified). Eleven months after exposure, lung tumor incidence was 49% (33/67) in the exposed animals and 27% (21/77) in the controls ($p < 0.01$).

In another study, Heston (1953) found that subcutaneous injections of sulfur mustard (0.05 cc of a 0.05% solution at weekly intervals for 6 weeks, or 0.1 cc of a 0.1% solution in olive oil at 2-day intervals for a total of 6 doses) into the mid-dorsal region of mice (strains A, C3H, and C3Hf) resulted in injection-site tumors, whereas injections of vehicle alone did not induce tumor formation. Tumors occurring at the injection site included sarcomas, sarcomas neurogenic in origin, a rhabdomyosarcoma, papillomas, a squamous cell carcinoma, a hemangi endothelioma, and a mammary carcinoma.

McNamara et al. (1975) exposed SDW rats, ICR Swiss albino and A/J mice, rabbits, guinea pigs, and dogs to sulfur mustard vapors for varying exposure durations up to one year. The test animals were exposed to 0.001 mg/m³, 24 hr/day, 5 days/wk, or daily to 0.1 mg/m³ for 6.5 hr followed by 0.0025 mg/m³ for 17.5 hr, 5 days/week. In the rat study, 70 males and 70 females were exposed at each of the two concentrations, and 50 of each sex were maintained as controls. No tumors were observed in rabbits, guinea pigs, dogs, or mice; however, skin tumors were seen in the rats and these were considered to be the result of exposure to sulfur mustard. The rats were tested in two separate studies; a "toxicity study" in which the animals were exposed for up to 52 weeks and then followed for 6 months at which time they were sacrificed, and a "carcinogenicity study" in which the animals were exposed for varying times up to 21 months and then observed for varying periods of time before being sacrificed. In both studies, skin tumors occurred in animals exposed to the highest concentration, but not in those exposed to the lower concentration.

Of the tumors observed in the exposed animals, McNamara et al. (1975) considered basal cell and squamous cell carcinomas, trichoepitheliomas, and keratoacanthomas of the skin to be related to the sulfur mustard exposure. The incidence of these tumors is shown in Tables 14 and 15.

Table 14. Incidence of Skin Tumors^a in Rats: Toxicity Study, as adapted by USEPA (1991)							
Exposure duration (months)	Post-exposure (days)	Exposure Group					
		Control		Low exposure^b		High exposure^c	
		M	F	M	F	M	F
2	0		0/5	0/5	0/5	0/5	
3	0	0/5	0/5	0/5	0/5	0/5	0/5
4	0				0/5	0/5	
6	0	0/5	0/5	0/5	0/5		
8	0	0/5	0/5	0/5	0/5	0/5	0/5
12	0	0/5	0/5	0/5	0/5 ^d	0/5	0/5
12	70					4/4 ^e	
12	90	0/4	0/4 ^f	0/4	0/5	0/1	0/5
12	180	0/7	0/4 ^g	0/6	0/14 ^h	0/6	5/13 ⁱ

SOURCE: McNamara et al., 1975

^a Only squamous cell and basal cell carcinomas of the skin were considered by McNamara et al. (1975) to be possibly related to the sulfur mustard exposure, and only these types are included in the numerators.

^b 0.001 mg/m³ 24 hr/day, 5 days/wk

^c 0.1 mg/m³ for 6.5 hr followed by 0.0025 mg/m³ for 17.5 hr for each day of exposure, 5 days/wk

^d Subcutaneous fibroma in 1 animal

^e Squamous cell carcinoma of the skin in 4 animals

^f Squamous cell carcinoma of uterus in 1 animal

^g Subcutaneous fibroma in 1 animal; pulmonary adenoma in one animal

^h Papilloma of the skin in one animal

ⁱ Squamous cell carcinoma of the skin in 4 animals; basal cell carcinoma of the skin in 1 animal; thyroid adenoma in 1 animal

Table 15. Incidence of Skin Tumors ^a in Rats: Cancer Study, as adapted by USEPA (1991)							
Exposure duration (weeks)	Post-exposure (months)	Exposure Groups					
		Controls	Tumors ^d	Low ^b	Tumors ^d	High ^c	Tumors ^d
1	13			0/1			
1	15					0/1	A
1	21			0/4	B	0/4	
2	20			0/5		0/5	C
4	16			0/1		0/1	
4	20			0/4		0/5	
8	15	0/4		0/2		0/4	
8	17			0/1			
8	18			0/1	D		
12	12			0/2		4/5	3F,G
12	17			0/3	E		
26	14			0/4		3/4	3F
26	18			1/1	F		
39	11			0/3	E	4/4	4F, H
52	2					1/1	F
52	4					1/1	H
52	6					1/1	F
52	7					0/1	
52	10	0/22	E	0/17		3/14	3E,2F,I
52	17	0/1	E			0/1	E
52	18					4/4	F

SOURCE: McNamara et al., 1975; as adapted by USEPA, 1991

^a Data for both sexes pooled; only squamous cell (F) and basal cell (G) carcinomas, trichoepitheliomas (H) and keratoacanthomas (I) of the skin were considered by McNamara et al. (1975) to be possibly related to the sulfur mustard exposure, and only these types are included in the numerators.

^b 0.001 mg/m³ 24 hr/day, 5 days/wk

^c 0.1 mg/m³ for 6.5 hr followed by 0.0025 mg/m³ for 17.5 hr for each day of exposure, 5 days/wk

^d Number and types of tumors: A. subcutaneous lipoma; B. axillary lipoma; C. subcutaneous fibroma; D. astrocytoma; E. skin, fibroma; F. skin, squamous cell carcinoma; G. skin, basal cell carcinoma; H. skin, trichoepithelioma; I. skin, keratoacanthoma. At 0.1 mg/m³, a single trichoepithelioma (type H tumor) occurred in the same animal with a squamous cell carcinoma.

2.3.11 Summary of Biological and Toxicological Properties

This summary section focuses on those aspects of the biological and toxicological properties of sulfur mustard that may have special relevance for the development of airborne exposure limits. These are discussed in the order in which they are presented in the previous sections.

Mechanism of Action (see Section 2.3.1):

- At levels of exposure below those producing acute toxicity, the target of sulfur mustard is DNA. Bifunctional alkylation reactions result in the formation of DNA strand cross-links leading to the inhibition of mitosis. Monofunctional alkylation reactions leading to other genotoxic effects may be responsible for mutagenic and possibly carcinogenic effects.
- Rapidly dividing cells are likely to be the most susceptible to low-concentration, subsymptomatic exposures to sulfur mustard.
- Corneal stem cells in the pericorneal conjunctiva of the eye may be very sensitive to sulfur mustard vapors.

Absorption and Distribution and Metabolism (see Section 2.3.2):

- Sulfur mustard is readily absorbed through epithelial membranes.
- Following intravenous or percutaneous exposures, sulfur mustard is distributed throughout most of the body except the eye.
- Following severe exposures, sulfur mustard can inhibit myelopoiesis in the bone marrow and thereby lead to immuno-deficiencies.

Local vs. Systemic Effects (see Section 2.3.3):

- Local effects on the eyes, skin and respiratory tract can occur in the absence of systemic effects.
- Systemic effects are not likely to occur at exposure levels below those producing signs of acute toxicity.

Acute Toxicity (see Section 2.3.4):

- Local acute effects are dose and time dependent, but, at concentration levels approaching the threshold, the Ct (concentration multiplied by exposure time) producing a given effect generally increases with increasing length of the exposure duration.

- The eye is more sensitive than the respiratory tract, which is more sensitive than the skin.
- Acute exposures to the eye affect the conjunctiva as well as the cornea.
- Minimal acute effects on the human eye have been seen at a vapor concentration of about 0.1 mg/m³, and at Cts of 1-12 mg-min/m³.
- Based on limited data, humans appear to be approximately three times more sensitive than rabbits and two times more sensitive than dogs, in terms of the effects of sulfur mustard on the eyes.

Subchronic/Chronic Toxicity (see Sections 2.3.5 and 2.3.6):

- Insufficient human data are available to evaluate potential chronic effects from prolonged exposure to subsymptomatic vapor concentrations.
- Acute ocular effects have been reported in humans occupationally exposed to sulfur mustard, but no documented evidence is available demonstrating that such effects occurred following prolonged exposure to subsymptomatic concentrations.
- Ocular effects are the most sensitive indicator of exposure in laboratory animals. Dose-dependent ocular effects occurred in dogs, but not in rabbits, mice, rats, or guinea pigs.
- Ocular effects, observed in dogs 16+ weeks after continuous exposure to concentrations not causing acute effects, were limited to the cornea and included vascularization, pigmentation, opacity, pannus, and granulation. Absence of conjunctival involvement suggests that the effects were not due to an inflammatory response. Absence of hematological changes (i.e., leukopenia) indicative of systemic toxicity suggest that the ocular effects were due to direct contact of the agent with the eye.

Chronic and Delayed/Recurrent Effects from Acute Exposures (see Section 2.3.7):

- Occupational exposures, with episodes of acute toxicity, can result in chronic bronchitis.
- Delayed and recurrent keratitis can occur 8-40 years after a severe vapor exposure.
- Sulfur mustard-induced immunosuppression may result in greater susceptibility to infections.

Developmental and Reproductive Effects (see Section 2.3.8):

- Acute exposures resulting in systemic uptake may have effects on reproductive organs, including inhibition of spermatogenesis.
- Fetal anomalies have been observed in the offspring of laboratory rats dosed with sulfur

mustard during gestation at dose levels that also resulted in maternal toxicity.

Genotoxicity (see Section 2.3.9):

- Sulfur mustard is genotoxic, producing DNA cross links, mutations following replication or repair errors, chromosomal breaks, and chromosomal aberrations.
- Increased frequencies of somatic cell mutations, sister chromatid exchanges, and chromosome abnormalities have been reported in individuals occupationally exposed to sulfur mustard.
- Studies with rats indicate that subchronic inhalation or oral exposures can result in dominant lethal effects.

Carcinogenicity (see Section 2.3.10):

- Sulfur mustard is classified by IARC and the NTP as a human carcinogen.
- Neoplastic changes occur in epithelial tissues after exposures that cause acute injuries (burns and blisters to the skin).
- Occupational exposures to sulfur mustard are associated with an increased risk of respiratory tract and skin cancers.
- Increased incidence of skin tumors has been reported for rats exposed to sulfur mustard, but not for dogs, mice, rabbits, or guinea pigs. Respiratory tract tumors were not seen in any of the tested laboratory species.
- Acute nonlymphocytic leukemia has been reported in patients receiving nitrogen mustard; evidence is suggestive regarding a link between sulfur mustard exposure and acute nonlymphocytic leukemia.

2.4 Existing Toxicity Values for Sulfur Mustard

2.4.1 Oral Reference Dose

An oral Reference Dose of 0.007 $\mu\text{g}/\text{kg}$ body weight/day has been derived for sulfur mustard by Opresko et al. (1998) from data presented in the Sasser et al. (1989b) reproductive toxicity study in rats. In the Sasser et al. (1989b) study, dose-related lesions of the squamous epithelium of the forestomach (acanthosis and hyperplasia) occurred at all dose levels and in both sexes. The incidence and severity of acanthosis was 0/94 in the controls, 71/94 in the low-dose group, 89/94 in the mid-dose group, and 94/94 in the high-dose group. The lowest tested dose of 0.03 mg/kg/day was considered a LOAEL and was adjusted for a 7-day/week exposure protocol. The chronic RfD was derived from the adjusted LOAEL of 0.022 mg/kg/day by applying a total uncertainty factor of 3000; 10 for protection of

sensitive subpopulations, 10 for animal-to-human extrapolation; 3 for LOAEL-to-NOAEL extrapolation, and 10 for extrapolation from a subchronic to chronic exposure.

This derivation and value were reviewed by the COT Subcommittee on Chronic Reference Doses for Selected Chemical-Warfare Agents, which concurred with the critical study and end point selection as well as the total uncertainty factor (UF) and reference dose value suggested by Opresko et al (1998). However, the COT Subcommittee recommended a slightly different choice of individual uncertainty factors (UF_A of 3 and UF_L of 10)(NRC, 1999; Bakshi et al., 2000). This reference dose value has now been formalized as an Army-wide reference value to be used in environmental risk assessments (Martinez-Lopez, 2000).

2.4.2 Inhalation Unit Risk

Rosenblatt (1987, unpublished) used the Japanese war gas factory worker data presented by Wada et al. (1968) to estimate a cancer slope factor (q_1^*) of 0.16 per (mg/kg)/day for sulfur mustard. Rosenblatt applied a modifying factor of 10 to obtain an adjusted slope factor of 1.6 per (mg/kg)/day. From context, it appears that the modifying factor of 10 was incorporated to account for uncertainties in the raw data and assumptions (p.3; Rosenblatt, 1987). From Rosenblatt's estimate, USEPA (1991) calculated an inhalation unit risk of 4.6×10^{-4} per ($\mu\text{g}/\text{m}^3$) [$1.6 \text{ per (mg/kg)/day} \times (20 \text{ m}^3/\text{day} \times 1/70 \text{ kg}) \times 10^{-3} \text{ mg}/\mu\text{g}$].

U.S. EPA (1991) also derived a cancer inhalation unit risk for sulfur mustard based on the results of animal exposure studies conducted by McNamara et al. (1975, see Section 2.3.10.2); however, EPA emphasized that the studies of McNamara et al. (1975) contained deficiencies that made a quantitative analysis difficult. The studies were conducted in 1970 and did not conform to current standards of experimental protocol, and bias was likely in the assignment of animals to the test categories (USEPA, 1991). In addition, many of the exposures were very brief, included only a few animals, many of which were sacrificed (and some were replaced) before their capacity to develop late-appearing tumors could be fully tested (USEPA, 1991). Despite these shortcomings, EPA noted that the McNamara et al. data are the best available for estimating the carcinogenic potency of sulfur mustard. The authors of the EPA report analyzed two sets of data from the McNamara studies: data from the previously described toxicity and carcinogenicity studies (see Section 2.3.10.2, Tables 14 and 15). EPA's analysis of data from the toxicity study included only those animals killed after the minimum latency period for first tumor appearance (12 months exposure plus 70 days observation post-exposure, see Table 14) (U.S. EPA, 1991). The resulting incidence of tumors is shown in Table 16.

In the McNamara et al. (1975) toxicity study almost all the rats were observed for 6 months after the 12-month exposures ended; therefore, USEPA estimated that the daily average exposure for the 18-month study duration to be equal to 2/3 (12 months/18 months) of the nominal concentration for the 12-month exposure (USEPA, 1991). Therefore, for the group exposed to $0.1 \text{ mg}/\text{m}^3$ followed by $0.0025 \text{ mg}/\text{m}^3$, EPA estimated average concentration for 18 months as $0.0193 \text{ mg}/\text{m}^3$ ($19.3 \mu\text{g}/\text{m}^3$). For the group exposed to $0.001 \text{ mg}/\text{m}^3$, the average concentration for 18 months was calculated to equal $0.00067 \text{ mg}/\text{m}^3$ ($0.67 \mu\text{g}/\text{m}^3$). These data were analyzed using the GLOBAL 86 computer program (a multistage dose-response model) (Howe et al., 1986) to calculate the 95% upper bound estimate of the slope at low dose (q_1^* or unit risk). The resulting unit risk was 2.9×10^{-2} per $\mu\text{g}/\text{m}^3$ (USEPA, 1991). The unit risk was then adjusted for a less-than-lifetime study duration (24 months for the rat) by multiplying by the ratio of the lifespan of the rat to study duration to the third power [i.e., $(24 \text{ mo}/18$

mo)³]. The same data (including the times that the animals were sacrificed) were also subjected to time-to-tumor analysis using the WEIBULL 82 computer program (Howe and Crump, 1983). Empirically, the latency time estimated from the Weibull model was about 2 months, and the lifetime upper-bound unit risk was estimated to be 8.5×10^{-2} per $\mu\text{g}/\text{m}^3$ (USEPA, 1991). The data were not adjusted for discontinuous exposures even though the rats were exposed for only 5 days/week (McNamara et al 1975; USEPA 1991).

Table 16. Rat Skin Tumor Data^a from McNamara et al. (1975) Toxicity Study Used in EPA Quantitative Assessment			
Sex	Exposure Groups		
	Control	Low exposure^b	High exposure^c
Males	0/11	0/10	4/11
Females	0/8	0/19	5/18
Both sexes	0/19	0/29	9/29

SOURCE: USEPA, 1991, derived from data presented in Table 14.

^a Includes only data for rats living longer than the time for first tumor appearance (12 months exposure plus 70 days post-exposure)

^b 0.001 mg/m³ for 24 hr/day, 5 days/wk

^c 0.1 mg/m³ for 6.5 hr followed by 0.0025 mg/m³ for 17.5 hr per day, 5 days/wk

If the USEPA analysis of the McNamara et al. (1975) **toxicity** study is re-evaluated to account for discontinuous exposures (only 5 days/wk; see Section 2.3.10.2 and Table 14, 15, and 16), the estimated unit risk derived from the Global 86 multistage dose-response model is 9.6×10^{-2} per $\mu\text{g}/\text{m}^3$

EPA also analyzed the data from the McNamara et al. (1975) carcinogenicity study using the multistage dose-response model (USEPA, 1991). In this study, exposure duration was short for some groups, but each group was observed for 13 to 21 months. Thirty exposure patterns were reduced to 17 exposure groups based on lifetime average daily exposures assuming a lifespan of 24 months (Table 17).

The major assumption in this analysis was that total cumulative exposure, not the pattern of exposure, is the major determinant of toxicity. The 95% upper bound on the slope at low dose was 9.4×10^{-2} per $\mu\text{g}/\text{m}^3$ (USEPA, 1991), based on no adjustment for discontinuous exposure.

Table 17. Rat Skin Tumor Data from McNamara et al (1975) Cancer Study Used in EPA's Quantitative Assessment			
Exposure Duration (weeks)	Exposure Concentration^a	Lifetime^b average daily exposure ($\mu\text{g}/\text{m}^3$)	Incidence of skin carcinomas
Control	-	0.0	0/27
1	low	0.0096	0/5
2	low	0.0192	0/5
4	low	0.0385	0/5
8	low	0.0769	0/4
12	low	0.115	0/5
26	low	0.250	0/4
1	high	0.279	0/5
39	low	0.375	0/3
52	low	0.500	0/17
2	high	0.558	0/5
4	high	1.12	0/6
8	high	2.23	0/4
12	high	3.35	4/5
26	high	7.25	4/5
39	high	10.9	4/4
52	high	14.5	10/23

SOURCE: USEPA, 1991, derived from data of McNamara et al., 1975

^a Low exposure was $0.001 \text{ mg}/\text{m}^3$ 24 hr/day, 5 days/wk; high exposure was $0.1 \text{ mg}/\text{m}^3$ for 6.5 hr daily and $0.0025 \text{ mg}/\text{m}^3$ for the remaining 17.5 hr daily, 5 days/wk.

^b A 2-yr lifetime was assumed.

If the USEPA analysis of the McNamara et al. (1975) **carcinogenicity** study is re-evaluated to account for discontinuous exposures (only 5 days/wk; see Section 2.3.10.2 and Table 14, 15, and 16), the estimated unit risk derived from the Global 86 multistage dose-response model is 13×10^{-2} per $\mu\text{g}/\text{m}^3$.

USEPA recommended a unit risk of 8.5×10^{-2} per $\mu\text{g}/\text{m}^3$, derived from the Weibull time-to-tumor model, as the most reliable upper bound estimate of the carcinogenic potency of sulfur mustard for a lifetime exposure to sulfur mustard vapors. The Weibull model takes into consideration killing of animals after less-than-lifetime exposure (USEPA, 1991). The USEPA authors also considered the Weibull model to be the most suitable, because the exposures were long-term, the model adjusts for killing the test animals before a full lifetime of exposure, and the sample size was the largest obtainable from the McNamara et al. (1975) data. When adjusted for discontinuous exposures, analysis of the toxicity and carcinogenicity cohorts using the GLOBAL 86 model supports a range of unit inhalation risk estimates (9.6×10^{-2} per $\mu\text{g}/\text{m}^3$ to 13.0×10^{-2} per $\mu\text{g}/\text{m}^3$), slightly above the unit risk value of 8.5×10^{-2} per $\mu\text{g}/\text{m}^3$ recommended by the USEPA in 1991 (USEPA, 1991).

EPA noted that the dose-response estimates derived from the McNamara et al. (1975) study were highly uncertain because the study did not follow a standard protocol. In addition, too few animals were exposed and observed for a lifetime to give adequate sensitivity for detecting late-developing effects (USEPA, 1991). Furthermore, the uncertainty concerning the experimental conditions was too great to allow more confidence about the absolute carcinogenic potency. Because malignant tumors appeared only at the highest mustard concentration and only late in life, EPA concluded:

"Perhaps it may exert its carcinogenic activity secondarily through lifelong exposure to its cytotoxic or irritating effects. Under such circumstances, human exposures at low concentrations for limited times may entail much less risk than implied by the unit risk factor estimated from lifetime effects at higher doses. On the other hand, the lack of low-dose responses and early-appearing tumors in the McNamara data may be due simply to the inherent difficulty of detecting low-risk levels in experiments of reasonable size".

Risk estimates for sulfur mustard have also been developed using relative potency methods. Using the results of studies by Heston (1950, see Section 2.3.10.2) and Shimkin and McClelland (1949), USEPA determined that the relative potency of sulfur mustard to induce pulmonary tumors in strain A mice was equivalent to that of 3-methylcholanthrene (MC) (USEPA, 1991). Using data on MC and benzo(a)pyrene (BaP) reported by Stoner et al. (1984), EPA determined that the potency of MC was 10-13 times that of benzo(a)pyrene (BaP) in inducing lung tumors in strain A mice. Because the potency of sulfur mustard was considered to be the same as that for MC, EPA concluded that the unit risk for sulfur mustard would be 10-13 times the inhalation cancer unit risk for BaP in humans. The unit risk for BaP (3.29 per mg/m^3) was derived from the oral slope factor of 11.5 ($\text{mg}/\text{kg}/\text{day}$)⁻¹ [EPA's estimate of the slope factor for BaP in 1991] converted to a unit risk using the standard defaults of 20 m^3/day for ventilation rate and 70 kg for body weight of humans. The resulting inhalation unit risk estimate for sulfur mustard, based on the relative potency method, was 33-43 per mg/m^3 (3.3×10^{-2} per $\mu\text{g}/\text{m}^3$ to 4.3×10^{-2} per $\mu\text{g}/\text{m}^3$) (USEPA, 1991). The reader should please note that the BaP slope factor currently accepted by EPA is a range, from 4.5 to 11.7 ($\text{mg}/\text{kg}/\text{day}$)⁻¹ (IRIS, 2000). The geometric mean of this range is 7.3 ($\text{mg}/\text{kg}/\text{day}$)⁻¹.

Watson et al. (1989) used the Rapid Screening of Hazard (RASH) (Jones et al., 1988) approach to develop a best estimate for sulfur mustard cancer potency; the Watson et al. (1989) analysis considered sulfur mustard to be 1.3 times as potent as BaP. The RASH method has been validated as an acceptable method for estimating carcinogenic potency (Omenn et al., 1995). Recently, Gaylor (1998) derived a BaP oral slope factor of less than 1.2 per mg/kg/day using the results of laboratory studies conducted by Culp et al. (1998) and using doses adjusted by body weight to the power of 3/4. Gaylor (1998) applied the relative potency of 1.3 for BaP to sulfur mustard (Watson et al., 1989) to the slope factor for BaP and obtained an estimated carcinogen potency factor of 1.6 per mg/kg/day for sulfur mustard.

Gaylor (1998) also estimated sulfur mustard slope factors of 5.0 and 2.6 (mg/kg/day)⁻¹ using linear extrapolations from benchmark doses producing forestomach hyperplasia or forestomach lesions in rats (Sasser et al., 1989a, 1989b), and a slope factor of 5.3 (mg/kg/day)⁻¹ using a method based on the maximum tolerated dose (Gaylor and Gold, 1995). These values convert to inhalation unit risks of 1.4 x 10⁻³ per ug/m³, 7.4 x 10⁻⁴ per ug/m³, and 1.5 x 10⁻³ per ug/m³, respectively. Unit risk is further analyzed in the dose response analysis presented as Section 3.3.3 of this report. A summary and geometric mean of unit risk values obtained by different methods examined in this report is also presented in Section 3.3.3.

2.5 Existing Air Exposure Limits for Sulfur Mustard

2.5.1 General Population Exposure Limits (GPL)

The current general population control limit for sulfur mustard is 0.0001 mg/m³ for a 72 hr time-weighted average (TWA) (DHHS, 1988; DA, 1991, 1997a,b). The selection of a 72-hr averaging period was based on limitations of the analytical methods available at the time, and it was recommended by DHHS that the capability to monitor for 0.0001 mg/m³ with a 12-hr sampling time be developed.

The GPL of 0.0001 mg/m³ was based on the studies and recommendations of McNamara et al. (1975). McNamara et al. (1975) also recommended an 8-hr TWA of 0.00017 mg/m³, a 3-hr TWA of 0.00033 mg/m³, and a Ceiling Limit of 0.01 mg/m³. These values were “placed arbitrarily at 1/30 of the values for the healthy workers (see Section 2.5.2) because such people are a much less homogenous group as far as age span and health are concerned”. A Ceiling value of 0.003 mg/m³ for the general population was adopted by the Department of Defense (DoD, 1984) and the Department of the Army (DA, 1991, 1997a,b). However, it was indicated that the Ceiling value may be an average value over the minimum time required to detect the specified concentration.

In recommending that the 0.0001 mg/m³ 72-hr TWA be adopted as a general population control limit, DHHS (1988) did not include an estimate of cancer risk. It was stated that because sulfur mustard is a human carcinogen “lower levels of exposure are of potential concern”. It was also noted that even though “the data suggest that mustard agent is less potent than such other known human carcinogens as tobacco, radon, and chromates, the data do not permit an estimate of the carcinogenic potency or the exact degree of carcinogenic risk with confidence”. However, the conclusion was reached by DHHS that the proposed control limit “will amply protect a general population 1000 meters or more from a demilitarization site or transportation route” (DHHS, 1988).

2.5.2 Worker Population Exposure Limits (WPL)

The current worker control limit for sulfur mustard is 0.003 mg/m^3 for an 8-hr time weighted average (TWA) (DHHS, 1988; DA, 1991). This value was based on the studies and recommendations of McNamara et al. (1975). McNamara et al. (1975) had recommended an 8-hr/day, 5 day/wk TWA of 0.003 mg/m^3 ; an 8-hr TWA of 0.005 mg/m^3 , a 3-hr TWA of 0.01 mg/m^3 , a 6-min TWA of 0.3 mg/m^3 , and a Ceiling Limit of 0.4 mg/m^3 . McNamara et al. (1975) based these recommendations on evidence that exposure to a concentration of 0.001 mg/m^3 for 24 hr/day ($\text{Ct} = 1.4 \text{ mg-min/m}^3$), 5 days/wk, for one year did not result in any detectable adverse effects (local, systemic, pathological, mutagenic, teratogenic, or carcinogenic) in five species of animals tested (see Section 2.3.6.2 for a description of this study). The worker control limit was derived by McNamara et al. (1975) by adjusting the sulfur mustard concentration of 0.001 mg/m^3 from a 24 hr/day exposure to an 8 hr/day exposure (i.e. $0.001 \text{ mg/m}^3 \times 24/8 = 0.003 \text{ mg/m}^3$). No other adjustments were made for extrapolating the results of the animal studies to humans.

In recommending that the 0.003 mg/m^3 8-hr TWA be adopted as a worker exposure limit, DHHS (1988) did not include an estimate of cancer risk. It was stated that because sulfur mustard is a human carcinogen "lower levels of exposure are of potential concern". It was also noted that "Although the data suggest that mustard agent is less potent than such other known human carcinogens as tobacco, radon, and chromates, the data do not permit an estimate of the carcinogenic potency or the exact degree of carcinogenic risk with confidence". However, the conclusion was reached by DHHS that the proposed workplace exposure limits "appear to provide adequate protection for workers during the limited time of potential exposure prior to the completion of the Chemical Stockpile Demilitarization Program" (DHHS, 1988).

3. FINDINGS AND DISCUSSION

3.1 Exposure Data

3.1.1 Human Data

3.1.1.1 Epidemiological Studies

A number of epidemiological studies have been conducted on soldiers, laboratory researchers, and factory workers exposed to sulfur mustard. Several of these are discussed in Section 2.3.6 and 2.3.10, dealing with the chronic toxicity and the carcinogenicity of the agent. In general, such studies are not useful for establishing exposure limits because insufficient information is available on the extent and duration of the exposure; i.e., a concentration-response relationship can not be determined.

Furthermore, in the past, exposures under such circumstances may have been to multiple chemicals, and/or sufficiently high, at times, to result in acute signs of toxicity. The delayed, recurrent, or chronic disabilities resulting from short-term, symptomatic exposures may not be appropriate for identifying health problems that might result from long-term exposures to very low concentrations that do not cause immediate signs of toxicity. Nevertheless, these studies do show that, in addition to possible increased incidence of tumors (see Section 2.3.10), the two major categories of effects that may occur following occupational exposures to sulfur mustard are ocular changes, such as conjunctivitis; and respiratory tract problems such as chronic bronchitis (see Section 2.3.6).

3.1.1.2 Laboratory Studies

Several laboratory studies evaluated the effects of sulfur mustard on the eyes of humans (Reed, 1918; Reed et al., 1918; Guild et al., 1941; Anderson, 1942). The studies of Reed and Reed et al. were summarized by Reed (1920). In the initial phase of this study, Reed subjected himself to a 45-min exposure to 12 mg per 10,000 liters of air, equivalent to 1.2 mg/m^3 for 45 minutes, or a Ct of 54 mg-min/m^3 . Reed reported that he developed severe blepharospasm and photophobia, profuse lacrimation, pronounced rhinitis, and a severe conjunctival injection which persisted for about six days and was still noticeable a month later. Although no pulmonary symptoms were reported, Reed also suffered a severe skin reaction, resulting in exfoliation of the mucous membranes and skin of the upper half of the body.

In the second part of these studies, Reed exposed test subjects to concentrations of sulfur mustard ranging from 0.1 to 1.0 mg/m^3 (Reed 1918 and Reed et al., 1918). In some tests only nominal concentrations were reported, but it was estimated that the nominal concentrations were 60-70% of the analytical concentrations. In one series of tests (Reed et al., 1918) the subjects wore respirator canisters (not reported whether these contained charcoal) and nose clips, and each had one eye protected with a goggle. Some of these individuals were reported to have participated in previous studies (Reed et al., 1918) and may have become sensitized to mustard agent. The mustard was sprayed as a mixture of freshly prepared agent and "absolute alcohol", and the alcohol aerosol may have enhanced the ocular and percutaneous effects of the agent, thus making the mustard more effective at lower concentrations. The resulting lesions varied from a mild, but distinct conjunctival injection with no skin burns, to a very severe conjunctivitis with photophobia, and skin burns. Three of seven men exposed to a nominal concentration of 0.001 mg/L (1 mg/m^3), for time periods ranging from 5 to 45 minutes, exhibited skin burns; one had moderately severe ocular effects and two had mild ocular effects. Of seventeen men exposed to 0.0005 mg/L (0.5 mg/m^3) for 10 to 45 minutes, one exhibited a skin burn and six developed conjunctivitis. Of thirteen men exposed to 0.0001 mg/L (0.1 mg/m^3) for 10 - 30 minutes, none developed skin burns, but three showed slight, but distinct, conjunctivitis.

Guild et al. (1941) studied the effects of sulfur mustard vapors on human volunteers exposed to concentrations of 0.1 to 65 mg/m^3 for time periods ranging from 2 min to 24 hr. It was reported that exposures to high concentrations (65 mg/m^3) for short durations (2 minutes) were less effective than longer exposures to lower concentrations. Guild et al. (1941) also concluded that, at equivalent Cts, very long exposures (more than 20 hours) were less effective than shorter exposures to higher concentrations. The longest exposure durations were 420, 480, and 600 min for single exposures, and one 1440-min cumulative exposure resulting from three 8-hour exposures on successive days. Based on these data, Guild et al. (1941) concluded that if the concentration of sulfur mustard was kept below 0.06 mg/m^3 , men working 8-hour shifts (daily Ct $\leq 29 \text{ mg-min/m}^3$) should not experience more than "a very slight degree of conjunctival reaction".

Anderson (1942) investigated the effects of mustard vapor under hot and humid weather conditions. The exposures were conducted in a chamber in which the maximum temperature was $91 \text{ }^\circ\text{F}$ and the maximum relative humidity was 97%. It was reported that Cts resulting in a specific effect were relatively similar within the range of concentrations studied (1 to 20 mg/m^3), however, they were also somewhat lower under tropical test conditions. At Cts of 60 mg-min/m^3 or above, some "huskiness" or partial loss of voice was noted in some subjects two to three days after the exposures. The severity of eye effects was relatively dose-dependent and at the lowest test Ct of 12.5 mg-min/m^3 , three of the four test subjects exhibited only a band of fine injection across the exposed part of the bulbar conjunctiva, and the fourth exhibited a trace of angular conjunctivitis. Cts of 12 - 30 mg-min/m^3 were associated with

obvious conjunctivitis possibly causing minor degrees of irritation in a few cases; Cts of 30-60 mg-min/m³ resulted in definite palpebral and bulbar conjunctivitis accompanied in a small proportion of cases by slight edema and transient photophobia; and Cts of 60-75 mg-min/m³ were associated with widespread conjunctivitis accompanied by chemosis and photophobia. It was reported that a Ct of 100 mg-min/m³ or more would likely result in 100% casualties. In summarizing the results of his study Anderson (1942) listed a Ct of 12 mg-min/m³ as a threshold for demonstrable eye effects, i.e., a “mild angular conjunctivitis without symptoms”.

The results of the studies of Reed (1918), Reed et al. (1918), Guild et al., 1941, and Anderson (1942), arranged by increasing Ct value, are summarized in Table 18. Taken together, these studies indicate that Cts of 1 to about 12 mg-min/m³ (concentrations of 0.1 to about 1.0 mg/m³ and time periods of 5-30 min) caused no ocular effects in about half the test subjects, and produced only mild effects in all but one of the others (as mentioned above, the effects observed in the Reed studies may have been enhanced due to the fact that the individuals were exposed to an aerosol, and many of the subjects had been previously exposed which may have made them hypersensitive to sulfur mustard). As shown in Table 18, concentrations of 0.1 mg/m³ for 480 or 600 min (Cts of 48 and 60 mg-min/m³), one of 0.24 mg/m³ for 210 min (Ct of 50 mg-min/m³) and one of 0.06 mg/m³ for three daily 8-hr periods (Ct of 86 mg-min/m³) caused only mild effects. [One anomalous result indicated that a concentration of 0.23 mg/m³ for 420 min (Ct of 97 mg-min/m³) caused severe ocular effects in 4 individuals]. As noted by Papirmeister et al. (1991), the ocular effects resulting from either very short exposures (1-2 min) or very long exposures (24 hr) are, in general, less than would be predicted from the effects seen at similar Cts administered over time periods of 6 min to 8 hr.

Table 18. Effect of Sulfur Mustard Aerosols or Vapors on the Eyes of Humans					
Ct (mg-min/m³)	Conc. (mg/m³)	Expos. Time (min)	Total Number Tested	Effects	Ref^f
1	0.1 ^{a,g}	10	6	None	A
1.5	0.1 ^{a,g}	15	2	Slight injection (1/2)	A
1.5	0.1 ^{b,g}	15	1	None	A
3	0.1 ^{a,g}	30	5	Slight injection (1/5) Marked injection (1/5)	A
4.8	0.48 ^{b,g}	10	2	Conjunctivitis (2/2)	B
5	1.0 ^{a,g}	5	1	Conjunctivitis, photophobia (1/1)	A
5	0.2 ^{a,g}	10	5	Conjunctival injection (2/5)	A

Table 18. Effect of Sulfur Mustard Aerosols or Vapors on the Eyes of Humans					
Ct (mg-min/m³)	Conc. (mg/m³)	Expos. Time (min)	Total Number Tested	Effects	Ref^f
5.8	0.58 ^{b,g}	10	2	Conjunctivitis (2/2)	B
7	0.7 ^{b,g}	10	1	Conjunctivitis (1/1)	A
7.5	0.5 ^{a,g}	15	3	Slight conjunctivitis (1/3)	A
9	0.3 ^{b,g}	30	1	None	A
9.4	0.47 ^{b,g}	20	3	Conjunctivitis (1/3)	B
10	0.5 ^{b,g}	20	1	None	A
10	1.0 ^{a,g}	10	2	Slight conjunctivitis (1/2)	A
10.5	0.7 ^{b,g}	15	1	Slight conjunctivitis (1/1)	A
11	0.55 ^{b,g}	20	2	None	B
11.6	0.58 ^{b,g}	20	2	Conjunctivitis (2/2)	B
12	0.48 ^{b,g}	25	2	Conjunctivitis (1/2)	B
12.5	6.3 ^h	2	4	Slight injection (3/4) Trace conjunctivitis (1/4)	D
13	2.6 ^{b,g}	5	1	None	A
15	1.0 ^{a,g}	15	2	None	A
15	0.5 ^{a,g}	30	8	Conjunctivitis (1/8) Marked conjunctivitis (1/8) Severe conjunctivitis (1/8)	A
20	1.0 ^{a,g}	20	1	Severe conjunctivitis (1/1)	A
21	1.4 ^{b,g}	15	1	None	A
22.5	0.5 ^{a,g}	45	1	None	A
23.1	6.9 ^h	3.33	4	Conjunctival injection (3/4) Trace conjunctivitis (1/4)	D

Table 18. Effect of Sulfur Mustard Aerosols or Vapors on the Eyes of Humans					
Ct (mg-min/m³)	Conc. (mg/m³)	Expos. Time (min)	Total Number Tested	Effects	Ref^f
27.5	10 ^h	2.75	3	Mild injection, exposed sclera (2/3) Band of injection (1/3)	D
31.5	0.7 ^{b,g}	45	1	None	A
34	6.8 ^h	5	3	Injection, edema (1/3) Marked injection, edema (2/3)	D
38.1	12.7 ^h	3	3	Band of injection (3/3)	D
41.8	12.6 ^h	3.33	3	Marked effects (3/3)	D
42	1.4 ^c	30	4	Generalized effect (4/4)	C
43	4.3 ^{a,g}	10	1	Marked conjunctivitis (1/1)	A
44	11 ^h	4	3	Marked injection, exposed conjunctiva (3/3)	D
45	1.0 ^{a,g}	45	1	Very severe conjunctivitis, photophobia	A
45.6	7.6 ^h	6	4	Moderate band of injection (1/4) Conjunctivitis, involving lids (3/4)	D
48	0.1 ^c	480	4	Slight congestion (4/4) ^d	C
48.8	13 ^h	3.75	3	Moderate injection of lids and conjunctiva (3/3)	D
49.8	10.5 ^h	4.75	3	Marked injection of lids and exposed conjunctiva (3/3)	D
50	0.24 ^c	210	4	Slight degree of angular reaction (4/4) ^d	C
50	2.5 ^h	20	3	Moderate injection, exposed sclera (3/3)	D
53	10.6 ^h	5	2	Generalized injection of conjunctiva (2/2)	D
54.6	15.6 ^h	3.5	1	Band of injection, exposed sclera and conjunctiva (1/1)	D
55.1	5.8 ^h	9.5	4	Marked conjunctival injection (3/4) Intense conjunctival injection (1/4)	D

Table 18. Effect of Sulfur Mustard Aerosols or Vapors on the Eyes of Humans					
Ct (mg-min/m ³)	Conc. (mg/m ³)	Expos. Time (min)	Total Number Tested	Effects	Ref ^f
56	14 ^h	4	3	Marked conjunctival injection (3/3)	D
56.1	1.7 ^h	33	3	Band of conjunctival injection, exposed sclera (3/3)	D
58	2.9 ^h	20	3	Moderate conjunctival congestion (3/3)	D
60	0.1 ^c	600	4	Slight generalized reaction (4/4) ^d	C
60	30 ^c	2	3	Slight congestion (3/3) ^d	C
60.7	4.5 ^h	13.5	3	Moderate injection, exposed sclera (3/3)	D
63	1.4 ^c	45	4	Generalized effect (4/4)	C
65	6.5 ^c	10	2	Slight generalized conjunctival reaction (2/2) ^d	C
65	13.7 ^h	4.75	3	Conjunctival injection, edema (2/3) Severe conjunctival injection (1/3)	D
70	5.0 ^h	14	3	Marked conjunctivitis, edema (2/3) Intense conjunctival congestion (1/3)	D
70.2	15.6 ^h	4.5	2	Injection of lids, exposed sclera (1/2) Severe conjunctival injection, corneal hazing, photophobia (1/2)	D
70.5	4.7 ^h	15	3	Marked conjunctivitis, lids injected, edema, photophobia (3/3)	D
72	72 ^c	1	4	Slight congestion (4/4) ^d	C
75	30 ^c	2.5	3	Very slight congestion (3/3) ^d	C
80	320 ^c	0.25	3	Moderate conjunctival congestion (3/3) ^d	C
86	0.06 ^c	1440 ^e	4	Scarcely discernible reaction (4/4) ^d	C
90	30 ^c	3	3	Slight angular congestion (3/3) ^d	C
97	0.23 ^c	420	4	Severe conjunctivitis, slight chemosis, photophobia, blepharospasm (4/4)	C

Table 18. Effect of Sulfur Mustard Aerosols or Vapors on the Eyes of Humans					
Ct (mg-min/m ³)	Conc. (mg/m ³)	Expos. Time (min)	Total Number Tested	Effects	Ref ^f
99	16.5 ^c	6	2	Severe conjunctivitis (2/2) Photophobia (1/2)	C
105	70 ^c	1.5	3	Slight to moderate congestion (4/4) ^d	C
144	144 ^c	1	6	Conjunctivitis, conjunctival congestion (5/6) Severe effects, photophobia (1/6)	C

SOURCE: See Reference list below

^a Nominal concentration; actual concentration estimated by Reed et al. (1918) to be 60-70% of the nominal

^b Analytical measurement; hydrogen ion method

^c Measured concentration; method not reported by Guild et al. (1941)

^d Number of subjects affected not clearly identified

^e Three 8-hr exposures resulting in "scarcely discernible" effect

^f References: A=Reed, 1918; B =Reed et al., 1918; C=Guild et al., 1941; D=Anderson, 1942

^g Sulfur mustard sprayed as an aerosol mixture of agent and "absolute alcohol"

^h Analytical measurement; Gold-benzidine method

3.1.2 Animal Exposure Data

The only animal study containing vapor exposure-response data for sulfur mustard is that of McNamara et al. (1975). In that study (Section 2.3.6.2) mice, guinea pigs, rabbits, rats and dogs were exposed to either 0.001 mg/m³ (24 hr/day, 5 days/wk), or to 0.1 mg/m³ for 6.5 hr/day followed by 0.0025 mg/m³ for 17.5 hr), 5 days per week (TWA of 0.029 mg/m³) for up to one year. The only dose-related effects observed were ocular abnormalities in dogs and skin tumors in rats. Although 10 dogs were tested at each vapor concentration only 2 dogs in the low exposure group and 4 in the high exposure group were tested for 52 wk. Six dogs in each group were exposed for 16 or more weeks and 4 in each group were exposed for 32 weeks. At the higher exposure, the incidence of ocular effects in dogs was 4/6 at 16 weeks, and 4/4 at 32 and 52 weeks (see Table 9). No ocular effects were seen in the low-exposure group.

3.2 Developing Exposure Limits: A Traditional Approach

3.2.1 Methods for Deriving Exposure Limits for Noncancer Endpoints

The objective of traditional toxicological, non-cancer risk assessment is to identify a threshold dose below which adverse health effects are not expected to occur, or are extremely unlikely (NRC, 1993). Lehman and Fitzhugh (1954) proposed that an acceptable daily intake (ADI) could be calculated for contaminants in human food. That concept was endorsed by the joint FAO-WHO (Food and Agricultural Organization and World Health Organization) Expert Committee on Food Additives in

1961 and subsequently adopted by the joint FAO-WHO Meeting of Experts on Pesticide residues in 1962 (McColl, 1990). Formally, the ADI was defined as:

$$\text{ADI} = \frac{\text{NOAEL}}{\text{SF}}$$

The NOEL stands for the no-observed-effect level in toxicological studies (the highest exposure level at which there are no statistically or biologically significant increases in frequency or severity of effects between the exposed population and its appropriate control), and the SF represents a safety factor to allow for variations in sensitivity in the human population and for differences in sensitivity between humans and the experimental animals. These two sources of variation often have been accommodated through the use of a $10 \times 10 = 100$ -fold SF (see NRC, 1970).

The basic approach described above has been modified by EPA (see Cicmanec et al. 1996 for review); the ADI has been redefined as a reference dose (RfD), a concept which takes into account the uncertainty associated with such an estimate. Consequently, safety factors have been replaced with uncertainty factors (UFs) which are scientifically based-descriptors of the various areas of uncertainty associated with a specific chemical compound (Barnes and Dourson, 1988). Uncertainty factors are used to account for: 1) individual variability in human response to the chemical; 2) to extrapolate from animal data to humans using the default assumption that humans may be more sensitive than the test species; 3) to extrapolate from a LOAEL (lowest-observable adverse effect level) to a NOAEL; 4) to extrapolate from an experimental subchronic exposure to a projected chronic or lifetime exposure; and 5) to account for the possibility that the true NOAEL or LOAEL was not identified due to deficiencies in the available toxicological data. A complete database is considered by USEPA to include chronic exposure studies with two species, developmental toxicity studies in two species, and a multi-generation reproductive toxicity study (USEPA, 1994). In deriving an RfD a modifying factor (MF) can also be used to increase or decrease the overall UF, depending on the professional judgement of the individuals assessing the reliability of the experimental data and the appropriateness of the calculated RfD. A RfD is derived using the following formula:

$$\text{RfD} = \frac{\text{NOAEL}_{\text{ADJ}}}{\text{UF}_H \times \text{UF}_A \times \text{UF}_L \times \text{UF}_S \times \text{UF}_D \times \text{MF}}$$

where:

NOAEL _{ADJ}	=	No-observed-adverse-effect level (mg/kg), adjusted for a daily exposure.
UF	=	Uncertainty Factor
UF _H	=	To account for human variability in response and for possible sensitive human subpopulations, if such factors can not be evaluated from the experimental data
UF _A	=	To extrapolate from animal data to humans using the default assumption that humans may be more sensitive than the test species
UF _L	=	To extrapolate from a LOAEL to a NOAEL
UF _S	=	To extrapolate from a subchronic to chronic exposure under the assumption that the LOAEL and NOAEL will occur at a lower dose with increasing exposure duration
UF _D	=	to account for the possibility that the true NOAEL or LOAEL was not identified because the appropriate toxicity test was not conducted (i.e., reproductive or developmental).

MF = Modifying Factor to adjust for chemical-specific, or study-specific uncertainties not dealt with by the standard uncertainty factors.

As defined by EPA, an RfD is “an estimate (with an uncertainty spanning perhaps an order of magnitude or greater) of a daily exposure level for the human population, including sensitive subpopulations, that is likely to be without an appreciable risk of deleterious effects” (USEPA, 1989). An RfD is expressed in terms of milligrams of chemical ingested per kilogram body weight per day. A daily exposure at or below the RfD is not likely to be associated with health risks, but as the daily intake increases above the RfD, the probability that an adverse effect will occur also increases (Cicmanec et al. 1996).

Values of 1, 3, and 10 are typically used for each individual UF, and the MF can range from 0.1 to 10 (the default MF is 1). The rationale for the selection of a particular UF value is given in Table 19. As noted by Cicmanec et al. (1996), the choice of the appropriate UF or MF reflects case-by-case judgement by experienced risk assessors, and the magnitude of any composite UF is dependent on professional judgement for the total uncertainty in all areas. Overall uncertainty is lowest if an RfD is derived from chronic exposure data for humans; however, in many cases the only available human data are from acute exposure studies and the only available chronic or subchronic data are from animal studies. Thus, the use of animal toxicity data, a less than a chronic exposure period, and/or the identification of a LOAEL and not a NOAEL adds several levels of uncertainty to derivation of an RfD.

The default value of 10 that is used for each UF is a conservative estimate of the associated uncertainty, and, when multiple default values are used, the resulting RfD is considered to be an estimate of a dose that is likely to be without adverse effects even in sensitive individuals for a lifetime of exposure (Dourson et al., 1996). Whenever there are adequate supporting scientific data, chemical-specific UFs should be used in place of the default value of 10. Dourson et al. (1996) and Young et al. (1999) provide specific examples of cases where UF values less than 10 have been used in deriving RfDs.

For uncertainty factors associated with individual human response variability and interspecies variability, Renwick (1993) suggested that these factors can be divided into two subfactors, a toxicokinetics component (relating external dose to internal dose) and a toxicodynamics component (relating internal dose with effect, or target organ sensitivity). Renwick (1993) examined intra- and interspecies differences in toxicokinetics and toxicodynamics and concluded that the UF_H and UF_A can be divided into subfactors of 4 for toxicokinetics and 2.5 for toxicodynamics. The International Programme on Chemical Safety (IPCS, 1994) agreed with Renwick’s subdivision of the UF_A , but recommended that the UF_H be subdivided evenly into a toxicokinetics component (3.16) and a toxicodynamics component (3.16). Further analysis of a larger database on inter-individual variability in response led Renwick and Lazarus (1998) to conclude that the division of the UF_H into two equal components is generally valid, but that the toxicokinetics subfactor of 3.16 may not be adequate for all routes of elimination for all subgroups of the population. For chemicals in which toxicokinetics are not critical in determining the magnitude or extent of the response, a UF_H of 3.16 (for differences in toxicodynamics) may be adequate for defining the uncertainty associated with inter-individual variability. Similarly, if toxicokinetics is not relevant, a UF_A of 3.16 for interspecies differences in toxicodynamics may be sufficient for extrapolating from animals to humans.

Table 19. Application of Uncertainty Factors in RfD/RfC Derivations			
Uncertainty Factor	Value		
	10	3	1
UF _H Sensitive humans	Default, if no other data are available	If scientific data indicate that a UF of less than 10 would be protective of the most sensitive population, or if toxicokinetics or toxicodynamics is not relevant.	If key study was conducted on sensitive or health-compromised human population, or if both toxicokinetic and toxicodynamic factors are not relevant.
UF _A Animal to human	Default, used to adjust for differences in species sensitivity	If differences in physiological parameters suggest that humans are less than 10-fold more sensitive than the test species. In the case of an RfC, when respiratory system dosimetric adjustments are made.	If there is physiological, biochemical and/or toxicological evidence that humans are not more sensitive than the test species
UF _S Subchronic to chronic	Default, to account for the possibility that the same effect will occur at a lower concentration/dose with longer exposure durations	If the test data indicate that the observed effect does not increase with increase in exposure duration due to the development of physiological compensating mechanisms.	If the toxicity value is based on a subchronic study but additional chronic data are available indicating no additional effects following longer exposures
UF _L LOAEL to NOAEL	Default, to estimate the NOAEL in the absence of complete information on the dose-response curve	If the effect seen at the LOAEL is of low severity; e.g., enzyme changes or organ weight changes in the absence of signs of toxicity <u>or</u> if dose-response analysis indicate that the NOAEL should occur at a higher dose level than 1/10th the LOAEL	RfD/RfC based on NOAEL
UF _D Database	Default, if the data base is lacking in several critical areas, i.e., reproductive or developmental toxicity, or a multi-generation study	If the data base is lacking in one or more key studies, <u>but</u> supporting data (i.e., mechanism of action and/or studies on related chemicals) indicate that the endpoints would not be of concern.	If the data base consists of multiple toxicity studies in more than 1 species (as, chronic studies in 2 or more species), reproductive/developmental toxicity studies in two species, and one multi-generation study.

SOURCE: Cicanec et al., 1996 and Young et al., 1999

The EPA has adapted the oral RfD method for estimating inhalation reference concentrations (RfCs) (USEPA, 1994). The RfC methodology departs from that for an oral RfD by using dosimetric adjustments to scale animal exposure concentrations to human equivalent concentrations for particular sections of the respiratory tract. Dosimetric adjustments differ for vapors and particle/aerosols. When a dosimetric adjustment is made, an animal-to-human uncertainty factor (UF_A) of 3 is used to account for potential species differences in toxicodynamics, in the absence of specific data on the relative sensitivity

of humans and animals. A UF_A of 1 is used if there are data showing that humans are not more sensitive than the test species.

The RfC method is appropriate for developing chemical agent Airborne Exposure Limits for non-cancer endpoints in the general population. Because a standardized method for deriving occupational exposure limits has not been established by the Occupational Safety and Health Administration (OSHA), it has been recommended that a modified RfD/RfC approach also be used to derive AELs for workers potentially exposed to chemical agents. The modified approach takes into account differences between the general population and workers in terms of exposure frequencies, exposure durations, and inhalation rates, and also includes consideration of the healthy worker effect.

3.2.2. Exposure Limits for Cancer Endpoints

Consideration must also be given to the potential carcinogenic risks associated with exposures to any chemical that is a known or suspect human carcinogen. A distinction can be made, however, between an individual's risk of cancer at a specific exposure limit, and the population risk (where the population size is factored into the decision).

Congress and regulatory agencies have not enunciated comprehensive cancer risk goals. A single point that delineates acceptable from unacceptable risk has not been set. The Supreme Court has noted that cancer risks of less than one-in-one million (1×10^{-6}) are trivial, but has not defined the point at which the risk becomes unacceptable. Although cancer risks are expressed as a probability of occurrence during a lifetime, they might also be expressed in terms of life shortening or earlier occurrence. Depending on background tumor incidence and the mode of action of a carcinogen, "a risk of one in a million may only be associated with a reduction in the time to tumors of a few hours or a few days in a human population" (Gaylor, 2000).

The EPA has not promulgated a single acceptable level of individual carcinogenic risk. For the Superfund program, the Agency has indicated that "for known or suspected carcinogens, acceptable exposure levels are generally concentration levels that represent an excess upper bound lifetime cancer risk to an individual of between 10^{-4} and 10^{-6} ." However, Travis et al. (1987) found that in decisions concerning hazardous waste sites where the affected geographic area is small and where population risks are presumably also small, past regulatory decisions indicate that 10^{-4} was used as a *de minimis* risk level for these sites; a *de minimis* risk being an acceptable level that is below regulatory concern.

In evaluating the cancer risks associated with the proposed sulfur mustard incinerator program at Aberdeen Proving Ground, EPA (1991) considered the predicted air concentrations at the boundary fence and determined that the maximum individual excess cancer risk would be 1.4×10^{-7} . For an estimated exposed population of 200,000, EPA calculated that this risk level would result in less than one case (the calculated number was 0.0004) of excess cancer per year. This was considered to be a negligible risk to the general population.

With respect to occupational exposures, the Occupational Safety and Health Act of 1970 charged the National Institutes of Occupational Safety and Health to "describe exposure levels that are safe for various employment, including but not limited to the exposure levels at which no employee will suffer impaired health or functional capacities or diminished life expectancies as a result of work

experience.” NIOSH developed a carcinogen policy that recommended “No detectable exposure levels for proven carcinogenic substances.” However, in 1980, the Occupational Safety and Health Administration attempted to lower the benzene standard from 10 ppm to 1 ppm. The Supreme Court struck this down, noting that OSHA had failed to identify significant risk at the higher level, or how that risk would be reduced at the lower level. The Court argued that OSHA is obligated to regulate only “significant risks” and that without a risk assessment of some kind, OSHA could not know whether a substance posed a significant risk. The court did not give useful guidance as to what constituted significant risk.

Likewise, NIOSH changed its no detectable exposure levels for carcinogens to lowest feasible concentration. In 1997, they adopted the approach of quantitative recommended exposure levels for carcinogens. These levels are recommended based on animal and human data, and an assessment of what can feasibly be achieved by engineering controls and measured by analytical techniques.

Of the carcinogens that federal agencies have chosen to regulate by application of quantitative cancer risk estimates, the target “acceptable risk,” or the risk that is represented by the established regulatory exposure limit, is variable. OSHA regulates risk for asbestos at 2×10^{-2} ; arsenic at 8×10^{-3} ; and ethylene dibromide, ethylene oxide, and formaldehyde in the 10^{-3} range. This level of risk is also the target range for a number of emissions regulated under Clean Air Act provisions. Therefore, it would appear that cancer risks in the 10^{-3} range are considered acceptable by oversight agencies.

In general, OSHA considers 10^{-3} a threshold of significant risk (Rodricks et al., 1987; Graham, 1993), and the agency usually does not regulate lower risks because of feasibility limitations (Lohner, 1997). Some examples follow.

In the case of benzene, the OSHA 8-hr TWA standard is 1 ppm (Code of Federal Regulations Chapter 29, Part 1910). This exposure is equivalent to 3.24 mg/m^3 for 8 hr/day, 5 days/wk. which is equivalent to $771 \text{ } \mu\text{g/m}^3$ for a 24 hr/day, 7 day/wk exposure. The inhalation unit risk for benzene (which falls in the range of $2.2 \times 10^{-6} (\text{ } \mu\text{g/m}^3)^{-1}$ to $7.8 \times 10^{-6} (\text{ } \mu\text{g/m}^3)^{-1}$ (currently posted on EPA's Integrated Risk Information System (IRIS)). Therefore, the cancer risk at the current OSHA standard is 1.7×10^{-3} to 6.4×10^{-3} , or 1.7 to 6.4 cases per thousand [using the standard equation, Risk = Concentration x Unit Risk; i.e., $771 \text{ } \mu\text{g/m}^3 \times (2.2 \text{ to } 8.3 \times 10^{-6} (\text{ } \mu\text{g/m}^3)^{-1})$]. The current OSHA standards for vinyl chloride and inorganic arsenic are 1 ppm (2.60 mg/m^3) and $10 \text{ } \mu\text{g/m}^3$, respectively, and the inhalation unit risks are $8.4 \times 10^{-5} (\text{ } \mu\text{g vinyl chloride/m}^3)^{-1}$ and $4.3 \times 10^{-3} (\text{ } \mu\text{g As/m}^3)^{-1}$ [values from EPA Health Effects Assessment Summary Table (HEAST) and IRIS]. Cancer risk levels calculated from the OSHA standards (adjusted for a continuous exposure) and from the inhalation unit risk values are 5.2 per one hundred exposed individuals for vinyl chloride and 1.02 per hundred exposed individuals for inorganic arsenic.

In recommending that the 0.003 mg/m^3 8-hr TWA be adopted as a worker exposure limit for sulfur mustard, DHHS (1988) did not include an estimate of cancer risk. It was stated that because sulfur mustard is a human carcinogen “lower levels of exposure are of potential concern”. However, the conclusion was reached by DHHS that the proposed workplace exposure limits “appear to provide adequate protection for workers during the limited time of potential exposure prior to the completion of the Chemical Stockpile Demilitarization Program”.

3.3 Carcinogenicity Assessment of Sulfur Mustard

3.3.1 Hazard Characterization

The overall database for human epidemiologic studies for sulfur mustard has shown a causal relationship between exposure to sulfur mustard and cancer in humans. EPA concluded that human exposure to sulfur mustard is linked to lung cancer in U.S. World War I veterans and in war gas factory workers in Japan, Germany, and England (USEPA, 1991). IOM (1993) concluded that there is a causal link between exposure to sulfur mustard and respiratory tract cancer (nasopharyngeal, laryngeal, and lung). A recent follow up of the Japanese cohort (Yamakido et al., 1996) presented evidence that continues to support a causal association between exposure to sulfur mustard and respiratory tract cancer, particularly lung cancer. However, exposures to the Japanese cohort were high as evidenced by frequent signs of acute toxicity such as skin blistering (Nakamura, 1956).

IARC (1987b) evaluated the cancer data for sulfur mustard and concluded that the evidence based on human data was "sufficient" and the evidence based on animal studies was "limited"; the resulting analysis placed sulfur mustard in IARC *Group 1* (carcinogenic to humans). EPA concurred with IARC's conclusion (USEPA, 1991). Evaluation of the evidence for carcinogenicity according to EPA's 1986 cancer risk assessment guidelines (USEPA, 1986a) would place sulfur mustard in EPA *Group A* (carcinogenic to humans) based on sufficient evidence in humans and limited evidence in animals. The weight of evidence based on EPA's proposed cancer risk assessment guidelines (USEPA, 1996) would place sulfur mustard in the "*known human carcinogen by inhalation exposure*" category based on sufficient evidence in humans showing a causal association between exposure to sulfur mustard and respiratory tract cancer in humans. Sulfur mustard has not been adequately tested in animal models.

The airborne exposure study by McNamara et al. (1975) used exposure durations ranging from only 1 to 12 months, which is not sufficient for inducing late-developing lesions, and the study protocol and exposure conditions were not adequately described. In the oral studies by Sasser et al., 1989a, 1996), the test material was administered for only 13-24 weeks, which is too short for detecting late-developing lesions; the test material was also administered as a bolus, which reduces the weight given to forestomach tumors. However, the induction of carcinomas of the skin of rats in McNamara et al (1975) adds weight that sulfur mustard is associated with carcinogenesis in organs of contact. Overall, the animal studies are less than adequate for hazard characterization of sulfur mustard.

3.3.2. Mode of Action of Sulfur Mustard

The mode of action of sulfur mustard appears to be both linear and nonlinear. As a direct-acting bifunctional alkylating agent (IARC, 1987a), sulfur mustard is expected to interact with DNA and cause various genotoxic effects. The evidence has shown that sulfur mustard is indeed genotoxic in bacteria (Stewart et al., 1989), yeast (Kircher and Brendel, 1983), and mammalian cells (Crathorn and Roberts, 1965, 1966; Walker and Thatcher, 1968; Scott et al., 1974; Jostes et al., 1989) when tested with in vitro or in vitro/in vivo assays. Sulfur mustard is also genotoxic in mammalian systems in vivo as evidenced by induction of dominant lethals in rats after inhalation (Rozmiarek et al., 1973) or oral exposure (Sasser et al., 1990). Data from the Japanese poison gas workers showed an increased frequency of mutations and sister chromatid exchanges in peripheral lymphocytes of the poison gas workers compared with controls matched for age and smoking status (Yanagida et al., 1988; Shakil et al., 1993). The absence of benign skin lesions as a precursor to malignancy in rats exposed to sulfur

mustard in air is additional evidence suggesting that sulfur mustard acts by the linear mode (McNamara et al., 1975). Only one squamous cell papilloma of the skin was reported and this occurred in the group with the low air concentration; otherwise only carcinomas were reported. The absence of regenerative hyperplasia or papillomas at the high air concentrations suggests a linear mode of action for sulfur mustard. Overall, the results from genotoxicity and carcinogenicity suggest that the primary mode of action for skin carcinogenesis by sulfur mustard induction involves a genetic mechanism and is likely linear. Because somatic mutations were observed in workers with exposure to sulfur mustard (other chemical exposures were also documented), the likely mode of action for respiratory cancer in humans is also linear.

Sulfur mustard is a vesicant capable of causing tissue damage, which can lead to cell proliferation (hyperplasia). Sasser et al. (1996) observed minimal hyperplasia of the forestomach epithelium after treatment of rats with 0.3 mg/kg/day 90 days; marked acanthosis was observed in the majority of animals treated with 0.4 mg/kg/day for about 22-24 weeks (Sasser et al., 1989a). The longer treatment time also resulted in a few squamous cell papillomas of the forestomach (Sasser et al., 1989a). The two studies by Sasser et al. (1989a and 1996) shows a progression of the forestomach lesion from minimal hyperplasia to acanthosis, to papilloma. With a longer treatment time, the papillomas may have progressed to carcinoma. Therefore, if epithelial cell proliferation (which had a no-effect level) is a prerequisite for the development of forestomach neoplasms (in this case papillomas), then the mode of action is likely to be nonlinear.

3.3.3. Dose-Response Analysis

In EPA's 1991 assessment, a traditional approach was applied to the McNamara et al. (1975) data for the sulfur mustard toxicity and carcinogenicity studies; i.e., extrapolation of dose-response data to estimate the 95% upper bound estimate of the slope (unit risk values). The following values were obtained: 6.8×10^{-2} (linearized multistage, toxicity study), 8.5×10^{-2} (Weibull, toxicity study), and 9.4×10^{-2} per $\mu\text{g}/\text{m}^3$ (linearized multistage, carcinogenicity study). The geometric mean of these values is 8.1×10^{-2} per $\mu\text{g}/\text{m}^3$. In their assessment of the carcinogenicity data of McNamara et al., EPA included an animal with a keratoacanthoma (a benign lesion that does not progress to cancer) with the malignant lesions. Excluding this animal from the dose-response analysis results in a unit risk of 8.7×10^{-2} per $\mu\text{g}/\text{m}^3$ compared with 9.4×10^{-2} per $\mu\text{g}/\text{m}^3$ as reported by EPA. The recalculated value is similar to that using the Weibull method. EPA's dose-response analysis did not include an adjustment for discontinuous exposure; McNamara et al. (1975) exposed the animals for only 5 days/week. Adjusting the concentrations for discontinuous exposure and analyzing the data using the linearized multistage model, yields unit risk values of 9.6×10^{-2} per $\mu\text{g}/\text{m}^3$ for the toxicity study and 13×10^{-2} per $\mu\text{g}/\text{m}^3$ (keratoacanthoma included) or 12×10^{-2} per $\mu\text{g}/\text{m}^3$ (keratoacanthoma excluded) for the carcinogenicity study. These values are only slightly different from those calculated by EPA.

According to EPA's 1996 proposed cancer guidelines, mechanistic data that suggest a mode of action should be applied to dose-response assessments. The mode of action of sulfur mustard appears to be low-dose linear and, therefore, supports a linear dose-response model for extrapolation to lower dose. The 1996 cancer guidelines propose that dose-response data be modeled within the range of experimental data to a point of departure, the lower 95% bound on the dose associated with a 10% increased risk (LED_{10}). From the point of departure, a straight-line extrapolation approach is used to calculate the slope ($0.1/\text{LED}_{10}$). Using the linearized multistage model to calculate LED_{10} values from the McNamara studies yields unit risk values of 9.0×10^{-2} per $\mu\text{g}/\text{m}^3$ (toxicity study) and 12×10^{-2} per

$\mu\text{g}/\text{m}^3$ (carcinogenicity study). These values are similar to those calculated using the low-dose extrapolation method.

EPA did not use data reported by Sasser et al. (1989a) to derive an oral slope factor or inhalation unit risk for sulfur mustard. Recently, Gaylor (1998) analyzed the Sasser et al. (1989a) data in which forestomach lesions developed in Sprague-Dawley rats administered sulfur mustard in the diet for about 22-24 weeks. The oral slope factor was 2.6 per mg/kg/day (2.7 per mg/kg/day for current assessment). Extrapolating from an oral study to an inhalation unit risk is accompanied by a large degree of uncertainty. Because the doses were not based on surface area of the targets and the test material was administered as a bolus, the confidence in the extrapolation method is decreased. However, confidence in the extrapolation method may be increased because the target for each route of exposure is an organ of contact (the respiratory tract for inhalation exposure to humans and the forestomach for oral exposure to rats) and factors such as absorption rates, first-pass effects, distribution, and elimination are not involved in the extrapolation. Further, in this particular case, route-to-route extrapolation has been reduced to route-to-route dose conversion, and extrapolation from oral to inhalation exposure implies that sulfur mustard is equally potent by both routes of exposure.

Because the McNamara data are less than adequate for conducting a quantitative assessment, the relative potency method (sulfur mustard compared with MC and MC compared with BaP) has been used to derive unit risk values for sulfur mustard. Using data from short-term carcinogenicity studies (i.e., pulmonary tumor induction in strain A mice), EPA determined that sulfur mustard is 10 to 13 times more potent than BaP and Watson et al. (1989) determined that it is 1.3 times more potent than BaP. EPA (U.S. EPA, 1991) reported a unit risk of 3.3×10^{-2} to 4.3×10^{-2} per $\mu\text{g}/\text{m}^3$ for sulfur mustard based on an oral slope factor of 11.3 per mg/kg/day for BaP. The slope factor was converted to an inhalation unit risk using a default ventilation rate ($20 \text{ m}^3/\text{day}$ and body weight (70 kg) for humans (adjustment factor was $0.286 \text{ (mg/kg)/day per mg/m}^3$), and a relative potency values of 10-13. Watson et al. (1989) utilized a slope factor of 6.1 per mg/kg/day for BaP derived by EPA (USEPA, 1986b) from an inhalation study in hamsters. Applying a relative potency of 1.3 (range of 0.6 to 2.9) reported by Watson et al. (1989) to the slope factor, EPA (USEPA, 1991) derived an inhalation unit risk of 2.3 per mg/m^3 ($2.3 \times 10^{-3} \text{ per } \mu\text{g/m}^3$ with a range of 1.0×10^{-3} to $5.1 \times 10^{-3} \text{ per } \mu\text{g/m}^3$).

Rosenblatt (1987, unpublished) used the Japanese war gas factory worker data presented by Wada et al. (1968) to estimate a cancer slope factor (q_1^*) of 0.16 per (mg/kg)/day for sulfur mustard. Rosenblatt applied a modifying factor of 10 to obtain an adjusted slope factor of 1.6 per (mg/kg)/day. From context, it appears that the modifying factor of 10 was incorporated to account for uncertainties in the raw data and assumptions (p.3; Rosenblatt, 1987). From Rosenblatt's estimate, USEPA (1991) calculated an inhalation unit risk of 4.6×10^{-4} per ($\mu\text{g}/\text{m}^3$) [$1.6 \text{ per (mg/kg)/day} \times (20 \text{ m}^3/\text{day} \times 1/70 \text{ kg}) \times 10^{-3} \text{ mg}/\mu\text{g}$].

Gaylor (1998) utilized data from a dietary study reported by Culp et al. (1998) and derived a slope factor of 1.2 per mg/kg/day for BaP (animal to human scaling was based on body weight^{3/4}); applying the relative potency of 1.3 yields an oral slope factor of 1.6 per mg/kg/day for sulfur mustard. Using human default values ($0.286 \text{ mg/kg/day per mg/m}^3$) to convert the oral slope factor to an inhalation unit risk gives a value of 0.46 per mg/m^3 ($4.6 \times 10^{-4} \text{ per } \mu\text{g}/\text{m}^3$). However, if EPA's relative potency factor (10-13) is used, the inhalation unit risk would be 3.43-4.46 per mg/m^3 (3.43×10^{-3} to $4.5 \times 10^{-3} \text{ per } \mu\text{g}/\text{m}^3$). Reanalyzing the Culp et al. (1998) data using the GLOBAL 86 linearized multistage computer program yields an oral slope factor of 2.6 per mg/kg/day (defaults: mouse body weight = 0.03 kg, body weight scaling = 3/4 power; slope factor = $0.1/\text{LED}_{10}$). Multiplying this slope factor by the

relative potency (1.3, 10, or 13) and converting the slope factor to an inhalation unit risk gives values of 0.97, 7.4, and 9.7 per mg/m^3 , respectively (9.7×10^{-4} , 7.4×10^{-3} , and 9.7×10^{-3} per $\mu\text{g}/\text{m}^3$).

Another way of examining these data and estimates is to consider deriving a new estimate of relative potency (of HD to benzoapyrene) by first determining the midpoint of the USEPA (1991) range of 10 to 13 (midpoint = 11.5). The geometric mean of this midpoint and the relative potency of 1.3 developed by Watson et al (1989) is 3.9. From the recent study of Culp et al. (1998), it is already known that a slope factor of 1.2 per $\text{mg}/\text{kg}/\text{day}$ for BaP can be derived. The resulting estimate of unit risk for sulfur mustard is thus 3.9×1.2 per $\text{mg}/\text{kg}/\text{day}$, which equals 4.7 per $\text{mg}/\text{kg}/\text{day}$. By applying a default ventilation rate (20 m^3/day and body weight (70 kg) for humans (adjustment factor was 0.286 ($\text{mg}/\text{kg}/\text{day}$ per mg/m^3), the value of 4.7 per $\text{mg}/\text{kg}/\text{day}$ converts to 1.3×10^{-3} per $\mu\text{g}/\text{m}^3$ (D. Gaylor, personal communication, 20 July, 1999).

Gaylor (1998) also derived slope factors of 5.0 and 2.6 per $\text{mg}/\text{kg}/\text{day}$ using linear extrapolation from benchmark dose [forestomach hyperplasia or forestomach lesions in rats from data in Sasser et al (1989a, b)] and a method based on maximum tolerated dose, respectively. Converting the slope factors to inhalation unit risk gives values of 1.4 per mg/m^3 (1.4×10^{-3} per $\mu\text{g}/\text{m}^3$) and 0.74 per mg/m^3 (7.4×10^{-4} $\mu\text{g}/\text{m}^3$).

Gaylor and Gold (1995) observed for 139 animal carcinogens tested in the National Toxicology Program that carcinogenicity potency can be estimated by 0.74 divided by the maximum tolerated dose, expressed in terms of $\text{mg}/\text{kg}/\text{day}$. Sasser et al (1989a) reported significant body weight depression in rats administered 0.3 $\text{mg}/\text{kg}/\text{day}$ sulfur mustard for 90 days. No toxic effects were noted at 0.1 $\text{mg}/\text{kg}/\text{day}$. Hence, a dose of 0.2 $\text{mg}/\text{kg}/\text{day}$ might serve as a maximum dose in a 2-year study. With a maximum tolerated dose of 0.2 $\text{mg}/\text{kg}/\text{day}$ for 5 days per week, the average daily dose at the maximum tolerated dose of $0.2 \times (5/7) = 0.14$ $\text{mg}/\text{kg}/\text{day}$. From Gaylor and Gold (1995), an estimate of the carcinogenic potency is less than $0.74/0.14 = 5.3$ per $\text{mg}/\text{kg}/\text{day}$ of sulfur mustard. Using human default values (0.286 $\text{mg}/\text{kg}/\text{day}$ per mg/m^3), to convert the oral slope factor to an inhalation unit risk, gives a value of 1.5 per mg/m^3 (1.5×10^{-3} per $\mu\text{g}/\text{m}^3$).

The current oral slope factor for BaP in the EPA IRIS Substance file (URL address: <http://www.epa.gov/ngispgm3/iris/>) is a range of values from 4.5 to 11.7 per $\text{mg}/\text{kg}/\text{day}$; the geometric mean of this range is 7.3 per $\text{mg}/\text{kg}/\text{day}$, which is the value that will be used in subsequent calculations. Multiplying this oral slope factor by the relative potency values (1.3, 10, and 13) results in slope factor of 9.5, 73, and 95 per $\text{mg}/\text{kg}/\text{day}$ for sulfur mustard. The corresponding inhalation unit risk values are 2.7, 21, and 27 per mg/m^3 (2.7×10^{-3} , 2.1×10^{-2} , and 2.7×10^{-2} per $\mu\text{g}/\text{m}^3$, respectively).

Use of the geometric mean of 7.3 per $\text{mg}/\text{kg}/\text{day}$ for the oral slope factor of BaP in the EPA IRIS Substance file and the new estimate of relative potency of HD to benzoapyrene derived above (= 3.9), give a sulfur mustard unit risk estimate of 3.9×7.3 per $\text{mg}/\text{kg}/\text{day}$, or 0.8×10^{-2} per $\mu\text{g}/\text{m}^3$ (D. Gaylor, personal communication, 20 July, 1999).

Table 20 summarizes the most robust unit risk values obtained by the different methods described above. Risk estimates represent values derived from sulfur mustard data for animals exposed to airborne concentrations, values derived by route-to-route extrapolation of sulfur mustard data from gavage-treated animals, values derived from relative potency methods, and one value [4.6×10^{-4} per ($\mu\text{g}/\text{m}^3$)] based on non-quantified estimates of air concentrations in Japanese war gas factories during World War II.

Table 20. Inhalation Unit Risk Estimates for Sulfur Mustard (HD)		
Source or Reference	Data set/Method	Unit Risk (per $\mu\text{g}/\text{m}^3$)
This report	HD data, airborne exposure, toxicity study data of McNamara et al., 1975; linear dose-response model to estimate (0.1/LED ₁₀)	9.0×10^{-2}
This report	HD data, air borne exposure, carcinogenicity study data of McNamara et al., 1975; linear dose-response model to estimate (0.1/LED ₁₀)	12.0×10^{-2}
Gaylor, 1998	HD data, oral exposure, squamous cell papilloma data of Sasser et al. (1989a); linear extrapolation from benchmark dose	7.4×10^{-4}
Gaylor, 1998	HD data, oral exposure, hyperplasia data of Sasser et al. (1989b)	1.4×10^{-3}
Gaylor, 1998	HD data, oral exposure data of Sasser et al., 1989a; maximum tolerated dose method using analysis of Gaylor and Gold (1995)	1.5×10^{-3}
Gaylor, 1999 ^a	Relative potency (HD to BaP) geometric mean of 3.9 derived from RP estimate of 1.3 (Watson et al., 1989) and midpoint of RP range (=11.5) in USEPA (1991). GM of RP times BaP potency <1.2 per mg/kg-d (Culp et al., 1998) and oral to inhalation extrapolation (0.286 mg/kg/day per mg/m ³)	1.3×10^{-3}
Gaylor, 1999 ^a	Relative potency (HD to BaP) estimate of 3.9 as derived above, BaP potency of 7.3 per mg/kg-d (IRIS 2000) and oral to inhalation extrapolation (0.286 mg/kg/day per mg/m ³)	8.0×10^{-3}
U.S. EPA, 1991	Derived from estimated q ₁ * of 1.6 per mg/kg/d calculated by Rosenblatt (1987) from non-quantitative air concentration estimate in Japanese war gas factory atmosphere and available cancer incidence data for former factory workers (Wada et al., 1968; Yamada et al., 1957), and oral to inhalation extrapolation (0.286 mg/kg/day per mg/m ³).	4.6×10^{-4}
GEOMETRIC MEAN OF ALL VALUES^b		4.1×10^{-3} per $\mu\text{g}/\text{m}^3$

^a Personal communication received 20 July 1999 from D. Gaylor, Associate Director, Risk Assessment, National Center for Toxicological Research (USFDA), Jefferson, AR

^b Equivalent inhalation slope factor is approximately 14 per mg/kg/day

The geometric mean of all the values in Table 20 is 4.1×10^{-3} per $\mu\text{g}/\text{m}^3$. This summary value appears to be reflective of the current body of knowledge on the carcinogenicity of sulfur mustard agent (airborne concentrations).

3.4 Selecting the Critical Effect for a Noncancer Endpoint

3.4.1 Human Data

As discussed in Sections 2.3.3 and 2.3.4, the eyes are affected by lower vapor concentrations of sulfur mustard than any other tissue in humans. As indicated in Tables 2, 3 and 4, the threshold for ocular effects is expected to be less than or equal to $12 \text{ mg}\cdot\text{min}/\text{m}^3$, whereas that for respiratory effects was reported to be between 12 and $70 \text{ mg}\cdot\text{min}/\text{m}^3$. The Cts listed in Tables 2-4 are, in general, representative of relatively high exposure concentrations (i.e., 1 to $10 \text{ mg}/\text{m}^3$ or higher) and relatively short exposure times (i.e., 1-10 min). Therefore, for deriving exposure limits, the critical effect is considered to be ocular changes, as indicated by conjunctival injection or mild conjunctivitis.

The severity of ocular damage in humans following exposure to sulfur mustard has been evaluated in several laboratory studies (Reed, 1918, Reed et al., 1918; Guild et al., 1941; and Anderson, 1942). The results of these studies are summarized in Section 3.1.1. Comparing the data from these studies is complicated by the fact that there may have been differences in experimental protocol, analytical method, individual exposure histories of the test subjects, and inherent or induced sensitivity (due to prior exposures). Furthermore, in at least two cases (the studies of Reed, 1918 and Reed et al., 1918), it was reported that the sulfur mustard was sprayed into the exposure chamber as a mixture of freshly prepared agent and absolute alcohol; therefore, the individuals were subjected to an aerosol exposure, which may have produced a response pattern different from and more severe than that caused by a vapor exposure alone. In aerosol exposures, differences in aerosol particle size and/or the uneven dispersion of the agent in the exposure chamber from one experiment to another might have resulted in fluctuations in the severity of the responses observed. Therefore, in the studies described by Reed it can not be determined with a high degree of certainty that the reported concentrations were the actual concentrations to which the subjects were exposed. In addition, it was also reported that some of the test subjects in the studies of Reed (1918) and Reed et al. (1918) had been previously exposed to sulfur mustard; consequently, an unknown number of individuals may have become hypersensitized.

Notwithstanding the limitations mentioned above, the data from these studies were combined, and the reported effects at each exposure level were categorized as to severity, to provide some measure of the exposure-response relationship for sulfur mustard (Table 21). The results of these studies indicate that exposures to concentrations of $0.1 \text{ mg}/\text{m}^3$ or less, for extended periods of time will cause only mild, if any, effects on the eyes of humans. These data also support the estimated Ct threshold for ocular effects as being less than $12 \text{ mg}\cdot\text{min}/\text{m}^3$, but they also indicate that, at low exposure concentrations, the threshold is defined more by the concentration than the exposure time, since only mild ocular effects were reported at Cts ranging from 1 to $60 \text{ mg}\cdot\text{min}/\text{m}^3$. As noted by Guild et al. (1941), at sulfur mustard concentrations of $0.1 \text{ mg}/\text{m}^3$ and below, an increase in the exposure period does not increase the severity of the lesion.

Table 21. Exposure-Response Data for Ocular Effects in Humans								
Conc. (mg/m ³)	Exp. Time (min)	Ct (mg-min/m ³)	Total Number	Number Showing Effects				Ref
				None	Mild	Mod.	Severe	
0.06 ^c	1440 ^e	86	4		4 ^e			C
0.1 ^a	10	1	6	6				A
0.1 ^a	15	1.5	2	1	1			A
0.1 ^b	15	1.5	1	1				A
0.1 ^a	30	3	5	3	2			A
0.1 ^c	480	48	4		4 ^d			C
0.1 ^c	600	60	4		4 ^d			C
0.2 ^a	10	5	5	3	2			A
0.23 ^c	420	97	4				4	C
0.24 ^c	210	50	4		4			C
0.3 ^b	30	9	1	1				A
0.47 ^b	20	9.4	3	2	1			B
0.48 ^b	10	4.8	2		2			B
0.48 ^b	25	12	2	1	1			B
0.5 ^a	45	22.5	1	1				A
0.5 ^b	20	10	1	1				A
0.5 ^a	15	7.5	3	2	1			A
0.5 ^a	30	15	8	5	1	2		A
0.55 ^b	20	11	2	2				B
0.58 ^b	10	5.8	2		2			B
0.58 ^b	20	11.6	2		2			B
0.7 ^b	45	31.5	1	1				A
0.7 ^b	10	7	1		1			A

Conc. (mg/m ³)	Exp. Time (min)	Ct (mg-min/m ³)	Total Number	Number Showing Effects				Ref
				None	Mild	Mod.	Severe	
0.7 ^b	15	10.5	1		1			A
1.0 ^a	15	15	2	2				A
1.0 ^a	5	5	1			1		A
1.0 ^a	10	10	2	1	1			A
1.0 ^a	20	20	1				1	A
1.0 ^a	45	45	1				1	A
1.4 ^b	15	21	1	1				A
1.4 ^c	30	42	4		4			C
1.4 ^c	45	63	4		4			C
1.7 ^g	33	56.1	3		3			D
2.5 ^g	20	50	3		1	2		D
2.6 ^b	5	13	1	1				A
2.9 ^g	20	58	3			3		D
4.3 ^a	10	43	1		1			A
4.5 ^g	13.5	60.7	3		2	1		D
5.0 ^g	14	70	3				3	D
5.8 ^g	9.5	55.1	4			3	1	D
6.3 ^g	2	12.5	4		4			D
6.5 ^c	10	65	2		2			C
6.8 ^g	5	34	3			3		D
6.9 ^g	3.33	23.1	4		4			D
7.6 ^g	6	45.6	4			3	1	D
10 ^g	2.75	27.5	3		3			D
10.5 ^g	4.75	49.8	3		1	2		D

Conc. (mg/m ³)	Exp. Time (min)	Ct (mg-min/m ³)	Total Number	Number Showing Effects				Ref
				None	Mild	Mod.	Severe	
10.6 ^g	5	53	2			2		D
11 ^g	4	44	3			3		D
12.6 ^g	3.33	41.8	3		2	1		D
12.7 ^g	3	38.1	3		3			D
13 ^g	3.75	48.8	3			3		D
14 ^g	4	56	3			3		D
14.1	5	70.5	3				3	D
14.6 ^g	4.45	65	3			2	1	D
15.6 ^g	3.5	54.6	1			1		D
15.6 ^g	4.5	70.2	2			1	1	D
16.5 ^c	6	99	2				2	C
30 ^c	2	60	3		3			C
30 ^c	2.5	75	3		3			C
30 ^c	3	90	3		3			C
70 ^c	1.5	105	3			3		C
72 ^c	1	72	4		4			C
144 ^c	1	144	6			4	2	C
320 ^c	0.25	75	3		3			C

SOURCE: See footnote f below; categorization of effects made by authors of this report.

^a Nominal concentration; actual concentration estimated by Reed et al. (1918) to be 60-70% of nominal

^b Analytical measurement; hydrogen-ion method

^c Analytical measurement; method not reported

^d Number affected not clearly stated

^e Three 8-hr exposures, scarcely discernible effect

^f References: A = Reed (1918); B = Reed et al. (1918); C = Guild et al. (1941); D = Anderson (1942)

^g Analytical measurement; Gold-benzidine method

3.4.2 Animal Data

In the only available long-term animal vapor exposure study, McNamara et al. (1975) considered that the only sulfur mustard-related effects were ocular changes in dogs exposed to 0.1 mg/m³ and keratitis in rats exposed to 0.001 mg/m³. Keratitis was not observed in rats exposed to 0.1 mg/m³, nor was keratitis reported for any of the control or exposed rats in the McNamara et al (1975) carcinogenicity study (note that these populations were exposed to the same concentration regime as those animals in the McNamara et al chronic toxicity study). As part of the current evaluation, chi square and Pearson correlation analyses of all rat keratitis data in McNamara et al (1975) were performed. The results of these tests indicate no statistical differences between the exposed and control populations, and no correlation of rat keratitis incidence with agent exposure (see Section 2.3.6.2 and Table 8). The absence of a positive dose-response relationship and statistical significance precludes the use of rat keratitis data for deriving exposure limits. In dogs, the eye was the most sensitive target organ with toxic effects occurring possibly as early as 3 months after the initiation of the exposure to 0.1 mg/m³. The small number of test and control animals makes it difficult to fully evaluate the results of the McNamara et al. study. Information was not provided on the breed of dogs used or the age and sex of the test animals. The reported increase in the body weights of the dogs over the 12-month test period (i.e., from about 10 to 12 kg) suggests that the animals were relatively young and therefore not likely to be suffering from age-related diseases. The observed ocular lesions are consistent with the known properties of sulfur mustard. Therefore, the concentration of 0.1 mg/m³ (0.029 mg/m³ time-weighted average) can be considered a LOAEL and the lower concentration of 0.001 mg/m³ can be considered a NOAEL for ocular effects.

It is likely that the ocular effects seen in the dogs were caused by direct contact of the sulfur mustard vapors with the epithelial tissues of the eye, and not the result of absorption through the lungs followed by systemic uptake and distribution. Tissue distribution studies indicate that systemically absorbed sulfur mustard does not reach the eyes (see Section 2.3.2), and in acute toxicity studies on rabbits, in which the agent was administered subcutaneously or by intravenous injection, there was no evidence of ocular effects even at dose levels that produced systemic toxicity (Warthin et al., 1918; Papirmeister et al., 1991; see Section 2.3.4). In addition, no agent-related ocular changes were reported in rats dosed orally with sulfur mustard for up to 13 weeks, nor in rats or rabbits receiving daily intragastric doses of sulfur mustard during gestation days 6-10 (rats) or 6-19 (rabbits), even at dose levels that were maternally toxic (Sasser et al., 1989a; Hackett et al., 1987).

3.5 Airborne Exposure Levels (AELs) for Sulfur Mustard

3.5.1 AELs for Chronic Exposures

3.5.1.1 General Population AELs for Chronic Exposures (GPLs)

GPL Derived from Human Data. Chronic human exposure data are not available for sulfur mustard in which a dose-response function and a LOAEL or NOAEL are clearly defined. As noted in Section 3.4, the critical effects observed in the short-term human studies were ocular changes, including redness, congestion, and irritation of the cornea. The longest experimental exposure involved one 8-hr exposure per day for three consecutive days (total exposure time 1440 min) to a sulfur mustard concentration of 0.06 mg/m³ (Guild et al., 1941). This experimental protocol included a 16-hr recovery period after the first and second exposures. The next longest human test with sulfur mustard involved a

single 10-hr exposure to 0.1 mg/m³ (Guild et al., 1941). Four individuals were exposed and the only reported effect was a "slight generalized reaction". Comparison of the exposure concentrations for the 8 hr/day, 3-day test with the single 10-hr test shows that the latter results in a lower time-weighted weekly average:

$$\begin{aligned} \text{LOAEL of } 0.06 \text{ mg/m}^3 \times 8 \text{ hr/24 hr} \times 3 \text{ days/7days} &= 0.008 \text{ mg/m}^3 \\ \text{LOAEL of } 0.1 \text{ mg/m}^3 \times 10 \text{ hr/24 hr} \times 1 \text{ day/7days} &= 0.006 \text{ mg/m}^3 \end{aligned}$$

Because the 10-hr test yields a lower, more conservative LOAEL for a continuous exposure, it is more appropriate for deriving a GPL. It should be noted that the calculation of an adjusted LOAEL assumes a linear response pattern over the time periods involved and may, to some degree, lead to an overly conservative GPL if the response pattern is less than linear for extrapolations to such long time periods.

To derive a GPL, the adjusted LOAEL of 0.006 mg/m³ is used in the standard equation:

$$\text{GPL} = \frac{\text{LOAEL}_{\text{ADJ}}}{\text{UF}_H \times \text{UF}_A \times \text{UF}_L \times \text{UF}_S \times \text{UF}_D \times \text{MF}}$$

where:

LOAEL _{ADJ}	=	0.006 mg/m ³ (Lowest-observed-adverse-effect level (mg/m ³), adjusted for a continuous exposure)
UF	=	Uncertainty Factor
UF _H	=	3 (to account for individual human variability in response)
UF _A	=	1 (human data)
UF _L	=	3 (To extrapolate from a LOAEL to a NOAEL)
UF _S	=	10 (To extrapolate from a short-term to long-term exposure)
UF _D	=	1 (data base adequate)
MF	=	3 (Modifying Factor; to adjust for chemical-specific, or study-specific uncertainties not dealt with by the standard uncertainty factors)

Therefore:

$$\text{GPL} = \frac{0.006 \text{ mg/m}^3}{(3)(1)(3)(10)(1)(3)} = 0.00002 \text{ mg/m}^3$$

A total uncertainty factor of 300 was applied, accounting for protection of sensitive subpopulations (3), extrapolation from a minimal LOAEL to a NOAEL (3), extrapolation from a short-term to long-term exposure (10), and a modifying factor (3).

UF_H - A UF_H of 3 is used for protection of sensitive subpopulations, because consideration must be given to the possibility that some groups such as children or the elderly may be more sensitive to ocular irritants. For skin exposures, the site of exposures and the thickness of the skin, which may be thinner in children and females, may make these subpopulations more sensitive to the skin vesicant effects of sulfur mustard (IOM, 1993). Differences in skin pigmentation are not an issue, as "there are no good experimental data to support the concept that there are substantial differences in the cutaneous response of black or white skin to antigen or injury" (IOM, 1993). A full UF_H of 10 is not used for a direct contact irritant such

as sulfur mustard because it is unlikely that the differences in sensitivity between the average and most sensitive individuals will be as large as an order of magnitude. The level of response may vary with anatomical and physiological differences associated with the eye (i.e., corneal thickness, amount of tearing), but because the effect is not the result of systemic absorption of the agent, physiological factors such as rates of metabolism or enzyme activity (which would account for much of the intraspecies uncertainty) would not be relevant. In terms used by Renwick and Lazarus (1998), it can be said that intrahuman differences in toxicokinetics would not be expected to be relevant; however, variability in toxicodynamics may be significant (see Section 3.2.1). A similar position has been taken by the National Advisory Committee on Acute Exposure Guideline Levels for Hazardous Substances (AEGs) which states "In cases where the mode or mechanism of action is such that the response elicited by exposure to the chemical by different subpopulations is unlikely to differ, an intraspecies uncertainty factor of 3 fold is generally used. Typically this involves a direct acting mechanism of toxicity where metabolism is unlikely to play a major role" (NAC/AEGL, 2000). No guidance is provided by the Committee on the possibility of enhanced adverse effects in individuals wearing contact lens, and there are no experimental or animal data on sulfur mustard to make such a determination.

- UF_A - Application of this UF is not needed because human data were used.
- UF_L - A UF_L of 3 is used to extrapolate to a NOAEL because the effects were reported to be a "slight generalized reaction" and therefore, the endpoint can be considered a minimal LOAEL.
- UF_S - A UF_S of 10 is used to extrapolate from the short-term exposure period of 10 hr to potential long-term exposures. There is considerable uncertainty as to the potential for cumulative effects to the eye at low exposure levels. Short-term exposure studies are not normally used for deriving a chronic toxicity value.
- UF_D - The toxicity data base for sulfur mustard contains subchronic/chronic vapor exposure studies in 5 species, two developmental toxicity studies in different species, a multi-generation reproductive bioassay, and a standard subchronic oral toxicity study in one species. Reproductive and developmental toxicity studies on sulfur mustard involved both the oral and inhalation pathways (see Section 2.3.8.2). The key study identifies a toxic effect that is consistent with the vesicant properties of sulfur mustard and a target organ (the eye) that is generally considered to be the most sensitive organ in humans exposed to sulfur mustard vapors. Therefore, the data base is adequate for deriving a GPL for sulfur mustard, and the use of a UF_D of 1 is justified.
- MF - A Modifying Factor of 3 is used to accommodate additional uncertainties inherent to the use of acute exposure data and the small number of subjects.

NOTE: Although sulfur mustard is known to produce sensitization (lowered response threshold) and latent effects (i.e., effects that do not appear until hours to days after the exposure occurs), the overall weight of evidence indicates that sensitization and latent effects are unlikely at the low level of exposure represented by the estimated GPL of 0.00002 mg/m³. Guild et al. (1941), indicate in their report that the test subjects were observed for at least 24 hr; therefore, the response levels would have included any latency period less than 24 hr. Furthermore, because effects were observed in test populations at the exposure levels used to derive the GPL, latency is not considered to be a relevant issue in this derivation.

Although sensitization has been reported for skin exposures, and assumed to be possible for respiratory exposures, it has not been reported for ocular exposures. Furthermore, the evidence suggests that induced sensitization only occurs following exposures that produce noticeable effects. Whether induced sensitization would occur at a GPL of 0.00002 mg/m^3 cannot be shown conclusively from the available data. However, a concentration of 0.00002 mg/m^3 is about 1/5000th of the concentration of 0.1 mg/m^3 at which only mild ocular effects have been observed (Table 21); therefore, the available data indicate that the possibility of induced sensitization at these concentrations is likely to be quite low.

GPL Derived from Animal Data - Ocular Effects. In the only available long-term animal vapor exposure study, McNamara et al. (1975) considered that the only sulfur mustard-related effects were ocular changes in dogs exposed to 0.1 mg/m^3 and keratitis in rats exposed to 0.001 mg/m^3 . Rats exposed to the high sulfur mustard concentration did not show a statistically significant increase in keratitis. Keratitis was not reported for any of the control or exposed rats in the McNamara et al (1975) carcinogenicity study. As part of the current evaluation, chi square and Pearson correlation analyses were performed on all rat keratitis incidence data; results indicate no statistical differences between the exposed and control populations, and no correlation with agent exposure (see Section 2.3.6.2 and Table 8). Thus, the rat keratitis data cannot be used for deriving a GPL. In dogs, the eye was the most sensitive target organ with toxic effects occurring possibly as early as 3 months after the initiation of the exposure to 0.1 mg/m^3 . However, ocular effects were not observed in the dogs exposed continuously to 0.001 mg/m^3 ; therefore, this latter concentration can be considered a NOAEL for ocular effects in dogs.

In the McNamara et al. (1975) study, exposure of the eyes of the test animals to sulfur mustard can be considered a direct function of the agent concentration in the exposure chamber, as well as anatomical and physiological/behavioral factors (i.e., blinking rate and rate of removal of the agent; amount of time the animals were awake relative to the amount of time they were asleep with their eyes closed; and the amount of moisture in the eyes). Because of the lack of data on these latter factors, the only appropriate measure of exposure is the concentration of sulfur mustard in the chamber. Consequently, the human equivalent exposure would be the same concentration (i.e., there is no need to adjust for species-specific differences in body size, respiratory rate, lung surface area, etc.). However, the animals were exposed for only 5 days/week in the McNamara et al. (1975) study; therefore, the concentration must be adjusted to a continuous 7 day/week exposure by using the factor of 5/7 (i.e., $5/7 \times 0.001 \text{ mg/m}^3 = 0.0007 \text{ mg/m}^3$). This time-adjusted NOAEL can then be used in the standard equation for deriving a GPL:

$$\text{GPL} = \frac{\text{NOAEL}_{\text{ADJ}}}{\text{UF}_H \times \text{UF}_A \times \text{UF}_L \times \text{UF}_S \times \text{UF}_D \times \text{MF}}$$

where:

$\text{NOAEL}_{\text{ADJ}}$	=	0.0007 mg/m^3 (No-observed-adverse-effect level, adjusted for exposure time)
UF	=	Uncertainty Factor
UF_H	=	3 (to protect sensitive subpopulations)
UF_A	=	3 (to extrapolate from animals to humans)
UF_L	=	1 (NOAEL)
UF_S	=	1 (chronic exposure)
UF_D	=	1 (data base adequate)
MF	=	3 (Modifying Factor; to adjust for deficiencies in the study)

Therefore:

$$\text{GPL} = \frac{0.0007 \text{ mg/m}^3}{(3)(3)(1)(1)(1)(3)} = 0.00002 \text{ mg/m}^3$$

A total uncertainty factor of 30 was applied, accounting for protection of sensitive subpopulations (3), extrapolation from animals to humans (3), and application of a Modifying Factor of 3.

UF_H - An Uncertainty Factor of 3 is used for protection of sensitive subpopulations, because consideration must be given to the possibility that some groups such as children and the elderly may be more sensitive to ocular irritants. For skin exposures, the site of exposures and the thickness of the skin, which may be thinner in children and females, may make these subpopulations more sensitive to the vesicant effects of sulfur mustard (IOM, 1993). Differences in skin pigmentation are not an issue, as "there are no good experimental data to support the concept that there are substantial differences in the cutaneous response of black or white skin to antigen or injury" (IOM, 1993). A full UF_H of 10 is not used for a direct contact irritant such as sulfur mustard because it is unlikely that the differences in ocular sensitivity between the average and most sensitive individuals will be as large as an order of magnitude. The level of response may vary with anatomical and physiological differences associated with the eye (i.e., corneal thickness, amount of tearing), but because the effect is not the result of systemic absorption of the agent, physiological factors such as rates of metabolism or enzyme activity (which would account for much of the intraspecies uncertainty) would not be relevant. In terms used by Renwick and Lazarus (1998), it can be said that intrahuman differences in toxicokinetics would not be expected to be relevant; however, variability in toxicodynamics may be significant (see Section 3.2.1). A similar position has been taken by the National Advisory Committee on Acute Exposure Guideline Levels for Hazardous Substances (AEGs) which states "In cases where the mode or mechanism of action is such that the response elicited by exposure to the chemical by different subpopulations is unlikely to differ, an intraspecies uncertainty factor of 3 fold is generally used. Typically this involves a direct acting mechanism of toxicity where metabolism is unlikely to play a major role" (NAC/AEGL, 2000).

UF_A - Application of a full UF_A of 10 for animal-to-human extrapolation is not considered necessary. For a contact irritant, interspecies variability in toxicokinetics is unlikely to be relevant; however, differences in toxicodynamics may be significant (Renwick and Lazarus, 1998). In the McNamara et al. (1975) study, dogs, guinea pigs, rabbits, rats and mice were exposed to sulfur mustard vapors. Only the dogs exhibited adverse ocular effects, even though rabbits are generally considered to be the most sensitive species with respect to ocular irritation due to several unique traits including the absence of a recognizable Bowman's membrane, loose eyelids, ineffective tear draining and poor blink response (Gilman, 1982; Battista and McSweeney, 1965; Buehler and Newman, 1964). However, the ocular effects seen in the dogs exposed to sulfur mustard related to the cornea, and Beckley (1965) found that a liquid detergent (50% water, 12% alcohol, and the remainder an

alkylbenzene sulfonate plus foam builder and stabilizer) caused slightly greater corneal effects in dogs and slightly greater iridial and conjunctival effects in rabbits. As noted by Grant (1974, differences in responses of the cornea may be partially related to differences in anatomy as well as chemical composition of the corneas of the various species. It has been reported that the thickness of the cornea of dogs is similar to that in humans (0.5 mm) (see Marzulli and Simmon, 1971; Maurice and Giardini, 1951; Mishima and Hedbys, 1968). Furthermore, it has been estimated that human eyes are 3-4 times more sensitive to sulfur mustard than rabbit eyes (see review by Gates and Moore, 1946) but only about 2 times as sensitive as the eyes of dogs (Henry, 1991, see Section 2.3.4.1). Dogs are therefore considered to be a better model than rabbits for the ocular effects of sulfur mustard because they are a more sensitive indicator of corneal injury. A UF_A of 3 was used to calculate a GPL because only species differences in toxicodynamics, and not toxicokinetics, are considered to be relevant, and the available data suggest that the eyes of humans are only 2 times more sensitive to sulfur mustard than the eyes of dogs.

- UF_L - An Uncertainty Factor of 1 is used because a NOAEL, not a LOAEL, is available. The same NOAEL of 0.001 mg/m^3 (time-adjusted to 0.0007 mg/m^3) was identified in five separate species. It should be noted that if a NOAEL was estimated from the higher sulfur mustard test concentration (TWA 0.029 mg/m^3) used in the McNamara et al. study, by applying the standard UF_L of 10, then the resulting NOAEL of 0.0029 mg/m^3 would be 3 times higher than that derived from the experimental data. This provides additional support for the use of 0.001 mg/m^3 as a NOAEL.
- UF_S - An Uncertainty Factor of 1 is used to extrapolate from a subchronic to chronic exposure. In the McNamara et al. (1975) study, the maximum exposure duration was 12 months. There are currently on IRIS oral RfDs for a least seven chemicals (butylate, benefin, captafol, carbofuran, cyanazine, heptachlor epoxide, and trifluralin) which were derived from 1-yr dog studies using a subchronic-to-chronic UF of 1 (USEPA, 1996). Therefore, there is a precedent for considering a duration of one year to be a chronic exposure.
- UF_D - The toxicity data base for sulfur mustard contains subchronic/chronic inhalation toxicity studies in 5 species, two developmental toxicity studies in different species, a multi-generation reproductive bioassay, and a standard subchronic oral toxicity study in one species. Reproductive and developmental toxicity studies on sulfur mustard involved both the oral and inhalation pathways (see Section 2.3.8.2). The principal study identifies a toxic effect that is consistent with the vesicant properties of sulfur mustard and there is no evidence that any other experimental species, including rabbits, is more sensitive to the agent. Furthermore, ocular effects are considered to be the most sensitive target organ in humans exposed to sulfur mustard vapors (McNamara et al., 1975). Therefore, the data base is adequate for deriving a GPL for sulfur mustard, and the use of a UF_D of 1 is justified.
- MF - A Modifying Factor of 3 is used because of deficiencies in the experimental protocol used in the critical study. Of particular importance is the small number of dogs (2 in the low exposure group and 4 in the high exposure group; gender not reported) exposed to sulfur mustard for a full one-year period. Current EPA guidelines for chronic toxicity testing with dogs recommend that four animals of each gender be used per dose level and for the concurrent controls (USEPA, 1998). Although an insufficient number of dogs were tested in the McNamara et al. (1975) study to meet modern-day experimental protocols, an adequate

number of rats (140), mice (140), rabbits (12) and guinea pigs (30) were tested: none of these other species exhibited any signs of ocular injury at the higher sulfur mustard concentration.

GPL Derived from Animal Data - Pulmonary Effects. Although there are no clear dose-response data for respiratory tract effects of sulfur mustard, chronic occupational exposures incurred by war gas factory workers, and acute combat exposures, are known to adversely affect the respiratory tract. Such exposures result in acute injuries and are associated with cases of non-malignant respiratory disease (IOM, 1993). Therefore, for the purposes of comparison with the GPL derived from the ocular effects endpoint, a GPL is derived here based on the potential pulmonary effects in rats exposed to HD (McNamara et al., 1975). The assumption used in this case is that, based on the McNamara et al. (1975) study, a reasonable NOAEL for pulmonary effects in rats is 0.001 mg HD/m³. The GPL is derived in accordance with current USEPA guidance (USEPA, 1994) for developing a Reference Concentration (RfC) for Category I type gases. Category I type gases are defined as "gases that are highly water soluble and/or rapidly irreversibly reactive in the respiratory tract" (USEPA, 1994:4-46). Although not highly water soluble, sulfur mustard fits the definition of being rapidly irreversibly reactive, and is thus considered a Category 1 gas.

The method used by EPA to derive an RfC for a Category I gas is to estimate a regional gas dose ratio for the region of the respiratory tract affected (RGDR_{PU}) for humans and the experimental species, using information on differences in lung surface area, minute volume, and mass transfer coefficients. The RGDR_{PU} for humans and rats was determined to be 2.23 (Major, 2000).

The RGDR_{PU} is then used as a dosimetric adjustment factor (DAF) to derive the human equivalent concentration (HEC) for the given effect level:

$$\text{NOAEL}_{\text{HEC}} = \text{NOAEL}_{\text{ADJ}} \times \text{DAF}$$

NOAEL_{ADJ} is the experimentally derived NOAEL from the animal study adjusted for the appropriate exposure frequency and duration. In the McNamara et al. (1975) study rats were exposed 24 hr per day, 5 days per week, to 0.001 mg/m³. Therefore, the NOAEL_{ADJ} for a continuous 7 day per week exposure is:

$$\text{NOAEL}_{\text{ADJ}} = 0.001 \text{ mg/m}^3 \times \frac{5}{7} = 0.0007 \text{ mg/m}^3$$

NOAEL_{HEC} is therefore derived as:

$$\text{NOAEL}_{\text{HEC}} = 0.0007 \text{ mg/m}^3 \times 2.23 = 0.0016 \text{ mg/m}^3$$

The GPL is derived by applying the appropriate Uncertainty and Modifying Factors:

$$\text{GPL} = \frac{0.0016 \text{ mg/m}^3}{(10)(3)(1)(1)(1)(3)} = 0.00002 \text{ mg/m}^3$$

where:

NOAEL _{HEC}	=	0.0007 mg/m ³
UF _H	=	10 (protection of sensitive subpopulations)
UF _A	=	3 (dosimetric adjustment used; therefore, 10 not required)
UF _L	=	1 (NOAEL used)
UF _S	=	1 (chronic study used)
UF _D	=	1 (data base complete)
MF	=	3 (deficiencies in study)

A total uncertainty factor of 100 was applied, accounting for protection of sensitive subpopulations (10) and animal to human extrapolation (3). A Modifying Factor of 3 was also applied to account for deficiencies in the experimental protocol used in the McNamara et al. (1975) study.

3.5.1.2 Worker AELs for Chronic Exposures (WPL)

An AEL for occupational exposures can be derived from the human studies conducted by Guild et al. (1941), and from the animal studies conducted by McNamara et al. (1975).

WPL Derived from Human Data. The longest intermittent exposure study for humans involved one 8-hr exposure per day for three consecutive days (total exposure time 1440 min) to a sulfur mustard concentration of 0.06 mg/m³ (resulting Ct of 86 mg-min/m³) (Guild et al., 1941). Four individuals were tested and it was reported that the effects of the agent on the eyes were "scarcely discernible". The exposure concentration of 0.06 mg/m³ for 8 hr/day for 3 consecutive days can be adjusted to a 5-day/wk occupational exposure by using a factor of 3/5. The resulting adjusted LOAEL is 0.036 mg/m³. This process assumes a linear response pattern over the time periods involved and may, to some degree, lead to an overly conservative WPL if the response pattern is less than linear. However, because the extrapolation is only from 3 days to 5 days, the potential error may not be significant. The adjusted LOAEL of 0.036 mg/m³ is used as follows:

$$\text{WPL} = \frac{0.036 \text{ mg/m}^3}{(1)(1)(3)(10)(1)(3)} = 0.0004 \text{ mg/m}^3$$

where:

LOAEL _{adj}	=	0.036 mg/m ³
UF _H	=	1
UF _A	=	1
UF _L	=	3
UF _S	=	10
UF _D	=	1
MF	=	3

A total uncertainty factor of 100 was applied; 3 for extrapolating from a LOAEL to a NOAEL, 10 for extrapolating from a short-term human exposure to a long-term exposure, and 3 as a Modifying Factor. Note that an uncertainty adjustment of (10) is the product of a UF of 3 and an MF of

3 which represent logarithmic means (3.16) of these order-of-magnitude factors. Hence, $3.16 \times 3.16 = 10$.

- UF_H - Adjustment for protection of sensitive subpopulations, such as children and the elderly or ill, is not considered necessary for a healthy worker population. For skin exposures, the site of exposures and the thickness of the skin may make female workers, whose skin may be thinner than males, more sensitive to the skin effects of sulfur mustard (IOM, 1993). Differences in skin pigmentation are not an issue, as "there are no good experimental data to support the concept that there are substantial differences in the cutaneous response of black or white skin to antigen or injury" (IOM, 1993). Furthermore, there is no evidence that the eyes of female workers would be more sensitive to sulfur mustard than the eyes of male workers. Although a wide range of inherent sensitivities to sulfur mustard may be present in the adult working population, idiosyncratic responders or those whose response is not predictable, are not normally considered a separate sensitive subpopulation. Occupational exposure standards are not generally based on the protection of such individuals. There is also evidence that in some individuals exposure to sulfur mustard may induce sensitization to subsequent exposures. Such individuals would also be considered to be hypersensitive. Although such sensitivity has been reported for skin exposures, and assumed to be possible for respiratory exposures, it has not been reported for ocular exposures. Furthermore, the evidence suggests that induced sensitization only occurs following exposures that produce noticeable effects. Whether induced hypersensitization would occur at a WPL of 0.0004 mg/m³ cannot be shown conclusively from the available data. However, a concentration of 0.0004 mg/m³ is 1/250th of the concentration of 0.1 mg/m³ at which only mild ocular effects have been observed (Table 21); therefore, the available data indicate that the possibility of induced sensitization at these concentrations is likely to be quite low.
- UF_A - Application of this UF is not needed because human data were used.
- UF_L - A UF_L of 3, not 10, is used to extrapolate to a NOAEL because the conjunctival effects were reported to be "scarcely discernible" and therefore, the endpoint can be considered a minimal LOAEL.
- UF_S - A UF_S of 10 is used to extrapolate from short-term exposure data to potential long-term exposures.
- UF_D - The data base for sulfur mustard is considered to be adequate (see discussion in Section 3.5.1.1)
- MF - A Modifying Factor of 3 is used to accommodate additional uncertainties inherent to the use of acute exposure data, and the small number of subjects.

NOTE: Although sulfur mustard is known to produce latent effects (i.e., effects that do not appear until hours to days after the exposure occurs), the overall weight of evidence indicates that latent effects are unlikely at the low level of exposure represented by the estimated WPL of 0.0004 mg/m³. Guild et al. (1941), indicate in their report that the test subjects were observed for at least 24 hr; therefore, the response levels would have included any latency period less than 24 hr. Furthermore, because effects were observed in test populations at the exposure levels used to derive the WPL, latency is not considered to be a relevant issue in this derivation.

The human exposure data listed in Table 21 show that exposure levels of 0.1 mg/m³ and below cause few, if any, ocular effects for exposure periods lasting up to 10 hr. Guild et al. (1941), state that at such low concentrations an increase in the exposure period will not lead to an increase in the severity of the lesion. If 0.1 mg/m³ is considered a threshold for mild effects, and if this value is reduced to a estimated no-effect level of 0.01 mg/m³ (by applying the standard factor of 10), it is unlikely that cumulative effects would occur. Such an estimated no-effect level of 0.01 mg/m³ is 25 times larger than the WPL of 0.0004 mg/m³ derived from the single data point of three 8-hr exposures.

WPL Derived from Animal Data - Ocular Effects. As noted in Section 3.5.1.1, the McNamara et al. (1975) study provides the only long-term animal vapor exposure data on which to base a chronic toxicity value. In that study, no non-cancer adverse effects occurred in rats, rabbits, mice, or guinea pigs exposed to the high sulfur mustard concentration (keratitis was observed in rats exposed to the low sulfur mustard concentration but found in the current evaluation to not be associated with agent exposure [see Section 2.3.6.2 and Table 8]. Skin tumors occurred in rats exposed to the high sulfur mustard concentration). Ocular changes occurred in dogs exposed to 0.1 mg/m³, but not in those exposed to 0.001 mg/m³; 24 hr/day, 5 days/wk for up to one year; therefore, the latter concentration is considered a NOAEL in dogs. The human equivalent exposure for workers is derived from this concentration by adjusting the exposure from the experimental 120 hr/wk to a 40-hr work week (i.e., 120 hr/40 hr x 0.001 mg/m³ = 0.003 mg/m³). This time-adjusted NOAEL can then be used in the standard equation for deriving an WPL. As in the case of the GPL there is no need to adjust for species-specific differences in body size, respiratory rate, lung surface area, etc., because the identified critical effect is a result of direct contact of the agent with the eyes.

$$\text{WPL} = \frac{0.003 \text{ mg/m}^3}{(1)(3)(1)(1)(1)(3)} = 0.0003 \text{ mg/m}^3$$

where:

$$\begin{aligned} \text{NOAEL} &= 0.003 \text{ mg/m}^3 \\ \text{UF}_H &= 1 \\ \text{UF}_A &= 3 \\ \text{UF}_L &= 1 \\ \text{UF}_S &= 1 \\ \text{UF}_D &= 1 \\ \text{MF} &= 3 \end{aligned}$$

A total uncertainty factor of 10 was applied; a factor of 3 for extrapolating from animals to humans, and a modifying factor of 3 to account for deficiencies in the study. The total uncertainty adjustment (10) is the product of a UF of 3 and an MF of 3 which represent logarithmic means (3.16) of these order-of-magnitude factors; hence, 3.16 x 3.16 = 10.

UF_H - For sulfur mustard, protection of sensitive subpopulations is not considered necessary for a healthy worker population. For skin exposures, the site of exposures and the thickness of the skin, which may be thinner in females, may make female workers more sensitive to the skin effects of sulfur mustard (IOM, 1993); however, there are no good experimental data to support the concept that there are substantial differences in the cutaneous response of black or white skin to antigen or injury (IOM, 1993). Furthermore, there is no evidence that the eyes of female workers would be more sensitive to sulfur mustard than the eyes of male

workers. Although a wide range of inherent sensitivities to sulfur mustard may be present in the adult working population, idiosyncratic responders or those whose response is not predictable, are not normally considered a separate sensitive subpopulation. Occupational exposure standards are not generally based on the protection of such individuals. There is evidence that in some individuals exposure to sulfur mustard may induce sensitization to subsequent exposures. Such individuals would also be considered to be hypersensitive. Although such sensitivity has been reported for skin exposures, and assumed to be possible for respiratory exposures, it has not been reported for ocular exposures. Furthermore, the evidence suggests that induced sensitization only occurs following exposures that produce noticeable effects. Whether induced hypersensitization would occur at the WPL, cannot be shown conclusively from the available data. However, a WPL concentration of 0.0003 mg/m³ is 1/333th of the concentration of 0.1 mg/m³ at which only mild ocular effects have been observed (Table 21); therefore, the available data indicate that the possibility of induced sensitization at these concentrations is likely to be quite low.

- UF_A - Application of a full UF_A of 10 for animal-to-human extrapolation is not considered necessary. For a contact irritant, interspecies variability in toxicokinetics is unlikely to be relevant; however, differences in toxicodynamics may be significant (Renwick and Lazarus, 1998). In the McNamara et al. (1975) study, dogs, guinea pigs, rabbits, rats and mice were exposed to sulfur mustard vapors. Only dogs exhibited concentration-dependent ocular effects (rats exhibited keratitis at the low concentration but not at the high concentration; additional statistical tests performed in the current evaluation find that the observed keratitis incidence is not associated with agent exposure; see Section 2.3.6.2 and Table 8), even though rabbits are generally considered to be the most sensitive species with respect to ocular irritation due to several unique traits including the absence of a recognizable Bowman's membrane, loose eyelids, ineffective tear draining and poor blink response (Gilman, 1982; Battista and McSweeney, 1965; Buehler and Newman, 1964). However, the ocular effects seen in the dogs exposed to sulfur mustard involved the cornea, and Beckley (1965) found that corneal damage from some liquid detergents was most severe in dogs while iridial and conjunctival responses were greatest in rabbits. As noted by Grant (1974), differences in responses of the cornea may be partially related to differences in anatomy as well as chemical composition of the corneas of the various species. It has been reported that the thickness of the cornea of dogs (~0.5 mm) is similar to that in humans (Marzulli and Simmon, 1971; Maurice and Giardini, 1951; Mishima and Hedbys, 1968). Furthermore, it has been estimated that human eyes are 3-4 times more sensitive to sulfur mustard than rabbit eyes (see review by Gates and Moore, 1946) but only about 2 times as sensitive as the eyes of dogs (Henry, 1991, see Section 2.3.4.1). Dogs are therefore considered to be a better model than rabbits for the ocular effects of sulfur mustard because they are a more sensitive indicator of corneal injury. A UF_A of 3 was used to calculate a WPL because only species differences in toxicodynamics, and not toxicokinetics, are considered to be relevant, and it is unlikely that eyes of humans are more than 3 times as sensitive to sulfur mustard as the eyes of dogs.
- UF_L - An Uncertainty Factor of 1 is used because a NOAEL is available.
- UF_S - The one year duration of the McNamara et al. study is considered to be a chronic exposure, therefore a subchronic to chronic extrapolation is not necessary.

UF_D - See discussion under GPL (Section 3.5.1.1) to support the use of a UF_D of 1.

MF - A Modifying Factor of 3 is used, as discussed in Section 3.5.1.1.

WPL Derived from Animal Data - Pulmonary Effects. Although there are no clear dose-response data for the respiratory tract effects of sulfur mustard, chronic occupational exposures incurred by war gas factory workers, and acute combat exposures, are known to adversely affect the lungs. Such exposures result in acute injuries and are associated with cases of non-malignant respiratory disease (IOM, 1993). Therefore, for the purposes of comparison with the WPL derived from the ocular effects endpoint, a WPL is derived here based on potential pulmonary effects in rats. The assumption in this case is that, based on the McNamara et al. (1974) study, a reasonable NOAEL for pulmonary effects in rats is 0.001 mg HD/m³. The WPL is derived in accordance with current USEPA guidance (USEPA, 1994) for developing Reference Concentrations (RfC) for Category I type gases. Category I type gases are defined as "gases that are highly water soluble and/or rapidly irreversibly reactive in the respiratory tract" (USEPA, 1994:4-46). Although not highly water soluble, sulfur mustard fits the definition of being irreversibly reactive, and is thus considered a Category 1 gas.

The method used by EPA to derive an RfC for a Category I gas is to estimate a regional gas dose ratio (RGDR_{PU}) for the region affected for humans and the experimental species, using information on differences in lung surface area, minute volume, and mass transfer coefficients. The RGDR_{PU} for humans and rats was determined to be 2.23 (Major, 2000).

The RGDR_{PU} is then used as a dosimetric adjustment factor for the respiratory tract region (DAF) to derive the human equivalent concentration (HEC) for the given effect level:

$$\text{NOAEL}_{\text{HEC}} = \text{NOAEL}_{\text{ADJ}} \times \text{DAF}$$

The NOAEL_{ADJ} is the experimentally derived NOAEL from the animal study adjusted for the appropriate exposure frequency and duration. In the McNamara et al. (1975) study rats were exposed 24 hr per day, 5 days per week, to 0.001 mg/m³. Therefore, the NOAEL_{ADJ} for a 8 hr per day, 5 day per week exposure is:

$$\text{NOAEL}_{\text{ADJ}} = 0.001 \text{ mg/m}^3 \times \frac{24}{8} = 0.003 \text{ mg/m}^3$$

And the NOAEL_{HEC} is derived as:

$$\text{NOAEL}_{\text{HEC}} = 0.003 \text{ mg/m}^3 \times 2.23 = 0.0067 \text{ mg/m}^3$$

The WPL is derived by applying the appropriate Uncertainty and Modifying Factors:

Therefore:

$$\text{WPL} = \frac{0.0067 \text{ mg/m}^3}{(1)(3)(1)(1)(1)(3)} = 0.0007 \text{ mg/m}^3$$

where:

NOAEL _{HEC}	=	0.0067 mg/m ³
UF _H	=	1 (healthy worker population)
UF _A	=	3 (dosimetric adjustment used; therefore, 10 not required)
UF _L	=	1 (NOAEL used)
UF _S	=	1 (chronic study used)
UF _D	=	1 (data base complete)
MF	=	3 (deficiencies in study)

A total uncertainty factor of 10 was applied; 3 for animal to human extrapolation and a Modifying Factor of 3 to account for deficiencies in the experimental protocol used in the McNamara et al. (1975) study.

3.5.2 AELs for Acute Exposures to Workers

3.5.2.1 Short-term Exposure Limit (STEL)

The American Conference of Government Industrial Hygienists (ACGIH) defines a STEL as "a 15-min time weighted average (TWA) exposure which should not be exceeded at any time during the workday even if the 8-hour TWA is within the TLV®-TWA (threshold limit value). Exposures above the TLV-TWA up to the STEL should not be longer than 15 minutes and should not occur more than four times per day. There should be at least 60 minutes between successive exposures in this range. An averaging period other than 15 minutes may be recommended when this is warranted by observed biological effects" (ACGIH, 2000, p. 4).

In calculating a STEL for sulfur mustard, data from human toxicity studies can be utilized. Because the effects that occur at the lowest exposure levels are ocular effects, these are the most appropriate to use in setting a STEL. Dose-response data for ocular effects are summarized in Table 19. Because a STEL is intended to be protective of four 15-min exposures per day, data points closest to a 60-min exposure were considered to be the most appropriate to use; these are presented in Table 22.

As shown in Table 22, the studies conducted by Reed (1918) and Reed et al. (1918) resulted in the lowest observed adverse effect levels; however, Reed et al. (1918) state that 68% of their test population were previously classified as to skin sensitivity and 52% had been burned previously by sulfur mustard one or more times. Furthermore, the subjects were exposed to a sulfur mustard aerosol containing absolute alcohol which may have enhanced the ocular and percutaneous effects of the agent. Therefore, it is concluded that the Reed studies are inappropriate for deriving a STEL. The next highest LOAEL identified in Table 22, is a 30-min exposure to 1.4 mg/m³ from the study of Guild et al. (1941).

In the present analysis, several approaches have been explored in the derivation of a STEL. They are provided in detail below:

Table 22. Dose-response Data for Ocular Effects in Humans used to Evaluate a STEL					
Conc. (mg/m ³)	Expos. Time (min)	Conc. (mg/m ³) for 60 min	Total Number Tested	Effects	Ref. ^a
0.06 ^b	30	0.03	5	Slight conjunctival injection (1/5) Marked injection (1/5)	A
0.18 ^b	30	0.09	1	None	A
0.3 ^b	30	0.15	8	Conjunctivitis (1/8) Marked conjunctivitis (1/8) Severe conjunctivitis (1/8)	A
0.3 ^b	45	0.15	1	None	A
0.35 ^b	45	0.17	1	None	A
0.48 ^c	25	0.24	2	Conjunctivitis (1/2)	B
1.4 ^c	30	0.7	4	Generalized effect (4/4) ^d	C
0.6 ^b	45	0.3	1	Very severe conjunctivitis, photophobia	A
1.7 ^c	33	0.94	3	Band of conjunctival injection, exposed sclera (3/3)	D
1.4 ^c	45	1.05	4	Generalized effect (4/4) ^d	C

SOURCE: See Footnote a

^a References: A=Reed, 1918; B = Reed et al., 1918; C=Guild et al., 1941; D=Anderson, 1942

^b Reed et al. (1918) state that the actual sulfur mustards concentrations were "probably only 60-70% of nominal"; the 60% value is given in this table; sulfur mustard was sprayed as an aerosol mixture of agent and "absolute alcohol" which may have accentuated the effects. Furthermore, some of the test subjects had been previously exposed.

^c Concentration based on analytical measurement

^d Number of subjects affected not clearly identified; assumed to be all tested

Minimal LOAEL Approach: The first approach is to consider the value of 1.4 mg/m³, derived from Guild et al. (1941) as a minimal LOAEL, and to calculate a STEL using the formula:

$$\text{STEL} = \text{LOAEL}_{\text{inhal}} \times \frac{\text{Resp}_{\text{exptl}} \times \text{Exp}_{\text{exptl}}}{\text{Resp}_{\text{occup}} \times \text{Exp}_{\text{occup}}} \times \frac{1}{\text{UFs} \times \text{MF}}$$

where: LOAEL_{inhal} = Lowest-observed adverse effect level for an inhalation exposure
 Resp_{exptl} = Respiratory rate of experimental population
 Resp_{ccup} = Respiratory rate for occupational exposures
 Exp_{exptl} = Exposure time period for experimental population
 Exp_{ccup} = Exposure time period for occupational exposures
 UFs = Uncertainty Factors (see Section 3.2.1)
 MF = Modifying Factor (see Section 3.2.1)

Because the reported effects are ocular and do not involve the respiratory tract, the adjustment for respiratory volume is not needed. Furthermore, as discussed in Section 3.5.2.1, the overall exposure-response data for ocular effects in humans supports a linear extrapolation over the time periods of concern for the STEL (i.e., from 30 min to 60 min). Therefore, the STEL can be estimated from the following relationship:

$$\text{STEL} = 1.4 \text{ mg/m}^3 \times \frac{30 \text{ min}}{60 \text{ min}} \times \frac{1}{10} = 0.07 \text{ mg/m}^3$$

This represents a NOAEL concentration adjusted for an exposure time totaling 60 min (i.e., up to four 15-min exposures in one day). The Uncertainty Factors used in the calculation are as follows:

UF_H = 1 (average human to sensitive human population)
 UF_A = 1 (animal to human extrapolation)
 UF_S = 3 (adjustment for the possibility of multiple exposures)
 UF_L = 3 (LOAEL to NOAEL extrapolation)
 UF_D = 1 (minimum to complete database)
 MF = 1 (not necessary)

A total uncertainty factor of 10 was applied:

UF_H - Protection of sensitive subpopulations is not considered necessary for a healthy worker population.

UF_A - Application of this UF is not needed because human data are used.

UF_S - A UF_S of 3 is used to extrapolate from a daily exposure at the STEL to possible multiple STEL exposures during the work week.

UF_L - A UF_L of 3 is used to extrapolate from a LOAEL to a NOAEL because the reported effects were described as a "generalized effect"; therefore, the endpoint is considered a minimal LOAEL.

UF_D - The data base for sulfur mustard is considered to be adequate (see discussion in Section 3.5.1.1).

MF - A Modifying Factor of 1 is used because no other uncertainties exist in the data.

The next highest LOAEL identified in Table 22, is a 33-min exposure to 1.7 mg/m³ from the study of Anderson (1942), which resulted in a band of conjunctival injection and exposed sclera. The value of 1.7 mg/m³ can also be used to calculate a STEL:

$$\text{STEL} = 1.7 \text{ mg/m}^3 \times \frac{33 \text{ min}}{60 \text{ min}} \times \frac{1}{10} = 0.09 \text{ mg/m}^3$$

This represents a NOAEL concentration adjusted for an exposure time totaling 60 min (i.e., up to four 15-min exposures in one day). The Uncertainty Factors used in the calculation are as follows:

UF _H	= 1 (average human to sensitive human population)
UF _A	= 1 (animal to human extrapolation)
UF _S	= 3 (adjustment for the possibility of multiple exposures)
UF _L	= 3 (LOAEL to NOAEL extrapolation)
UF _D	= 1 (minimum to complete database)
MF	= 1 (not necessary)

Time-Adjusted LOAEL approach: It is acknowledged that the available data from which to estimate a STEL do not fully address the time periods of interest. It is further acknowledged that there is as preference for using available human data, with appropriate precautions. For reasons previously described, the studies of Reed (1918, 1920), Reed et al (1918) and Anderson (1942) are considered inappropriate for use in deriving a STEL.

The database of human exposure to sulfur mustard includes the intermittent exposure study of Guild et al. (1941), in which adult males received one 8-hr exposure per day for three consecutive days (total exposure time 1440 min) to a sulfur mustard concentration of 0.06 mg/m³ (resulting Ct of 86 mg-min/m³) (Guild et al., 1941). Four individuals were tested and it was reported that the effects of the agent on the eyes were "scarcely discernible". The exposure concentration of 0.06 mg/m³ for 8 hr/day for 3 consecutive days can be adjusted to a 5 day/wk occupational exposure by using a factor of 3/5. The resulting adjusted LOAEL is 0.036 mg/m³. This process assumes a linear response pattern over the time periods involved and may, to some degree, lead to an overly conservative estimate if the response pattern is less than linear. The adjusted LOAEL of 0.036 mg/m³ is applied as follows:

$$\text{STEL} = \frac{0.036 \text{ mg/m}^3}{(1)(1)(3)(3)(1)(1)} = 0.0036 \text{ mg/m}^3$$

where:

LOAEL _{adj}	= 0.036 mg/m ³
UF _H	= 1
UF _A	= 1
UF _L	= 3
UF _S	= 3
UF _D	= 1
MF	= 1

A total uncertainty factor of 10 was applied; 3 for extrapolating from a LOAEL to a NOAEL, and 3 for extrapolating from a short-term human exposure. Note that an uncertainty adjustment of (10) is the product of two UFs of 3, which represent logarithmic means (3.16) of these order-of-magnitude factors. Hence, $3.16 \times 3.16 = 10$.

- UF_H - Adjustment for protection of sensitive subpopulations is not considered necessary for a healthy worker population.
- UF_A - Application of this UF is not needed because human data were used.
- UF_L - A UF_L of 3, not 10, is used to extrapolate to a NOAEL because the conjunctival effects were reported to be "scarcely discernible" and therefore, the endpoint can be considered a minimal LOAEL.
- UF_S - A UF_S of 3 is used to extrapolate from short-term exposure data
- UF_D - The data base for sulfur mustard is considered to be adequate (see discussion in Section 3.5.1.1)
- MF - A Modifying Factor of 1 is used

NOTE: Although sulfur mustard is known to produce latent effects (i.e., effects that do not appear until hours to days after the exposure occurs), the overall weight of evidence indicates that latent effects are unlikely at the low level of exposure represented by the estimated STEL of 0.0036 mg/m³. Guild et al. (1941), indicate in their report that the test subjects were observed for at least 24 hr; therefore, the response levels would have included any latency period less than 24 hr.

Probit and Logistics Approach: A third approach is to consider statistical analysis. Dose-response data for ocular effects in humans, categorized by severity of response (Table 21) were also used to calculate a STEL by means of two statistical procedures (probit and logistics analyses, see Appendix A). In both cases the concentration data for the Reed (1918) and Reed et al. (1918) studies were converted to 60% of the nominal values (Reed et al., 1918, estimated that the actual concentrations may have been 60-70% of nominal; the use of 60% for both data sets is a protective approach). Only the exposures resulting in (either or both) no effects or mild effects were used in the analyses. The time period was fixed at 60-min corresponding to that appropriate for a STEL, and the sulfur mustard concentration was calculated for the value at which no ocular effects would occur in 99% of the exposed population. The results are as follows:

Probit analysis: STEL = 0.0309 mg/m³
Logistics analysis: STEL = 0.0669 mg/m³

Since both statistical procedures predict the concentration at which 1% of the population might show an effect, to arrive at a NOAEL, an Uncertainty Factor of 3 can be applied to estimate the true NOAEL, and, furthermore, because of the potential for multiple STEL exposures occurring throughout the workweek, an additional Uncertainty Factor of 3 could be used for UF_S. The total Uncertainty Factor would therefore be 10, and the resulting STEL would fall in the range of 0.003 to

0.006 mg/m³. Thus, a STEL of 0.003 mg/m³ falls in the lower end of the calculated range and would be expected to be adequately protective.

STEL summary: Calculations of the STEL from human data for exposure periods of one hour or less resulted in STEL (four 15 min periods of exposure) values which, when averaged over an 8-hour workday, exceeded the proposed WPL 8-hr TWA of 0.0004 mg/m³ as follows: 0.07 mg/m³ for 1 hr averaged over 8 hours is 0.0088 mg/m³, while 0.09 mg/m³ for 1 hr averaged over 8 hr is 0.01 mg/m³. Note that four 15-min STELs of 0.003 mg/m³, averaged over 8 hr, without any other exposure, equals 0.000375 mg/m³.

Alternative approaches for estimating the STEL using 3-day consecutive exposure data from the Guild et al (1941) study (the "Time-Adjusted LOAEL approach") and a statistical analysis (the "Probit and Logistics Approach") indicate that a STEL value approximating 0.003 mg/m³ would be suitable and adequately protective.

As noted previously, the human experimental data indicate a threshold of about 0.1 mg/m³ for mild ocular effects (Table 21), regardless of the exposure time. A no-effect level might be estimated to be 0.01 mg/m³ using the standard default of 10. The estimated STEL of 0.003 mg/m³ is below the estimated no-effect level by an approximate factor of 3.

In addition, a STEL of 0.003 mg/m³ would be consistent with the current "alarm" value of 0.003 mg/m³ established by Army regulation (AR 385-61; DA 1997a) for mustard agent workers. This same air concentration is also the action level for mustard agent workers to don a "NIOSH/MSHA approved pressure demand, full-face piece, SCBA or supplied air respirator" (p. 6, AR 385-61; DA 1997a).

Therefore, for technical and operational reasons, the protective STEL estimate of 0.003 mg/m³ is recommended.

3.5.2.2 Immediately Dangerous to Life or Health (IDLH) Exposure Limit

The current NIOSH definition for an immediately dangerous to health or life condition (NIOSH Regulator Decision Logic, 1987) is a situation "that poses a threat of exposure to airborne contaminants when that exposure is likely to cause death or immediate or delayed permanent adverse health effects or prevent escape from such an environment." It is also stated that the purpose of establishing an IDLH is to "ensure that the worker can escape from a given contaminated environment in the event of failure of the respiratory protection equipment."

The most recently revised criteria (NIOSH Publication No. PB-94-195047), for determining an IDLH involve a tiered approach with:

1. acute human toxicity data being used preferentially followed next by
2. acute animal inhalation toxicity data (lethal concentration adjusted to an equivalent 30-min exposure, if necessary): lethal concentrations are divided by a safety factor of 10

3. acute animal oral toxicity data; the lethal dose is used to determine the equivalent total dose to a 70 kg worker and the air concentration containing this dose was determined by dividing by 10 cubic meters; IDLH was determined by dividing these air concentrations by a safety factor of 10, and
4. chronic toxicity data are considered if no relevant acute toxicity data exist although they may have little relevance to acute effects.

In the OSHA regulation (29 CFR 1910.146) on permit-required confined spaces, an immediately dangerous to life or health condition is defined as follows:

"Any condition that poses an immediate or delayed threat to life or that would cause irreversible adverse health effects or that would interfere with an individual's ability to escape unaided from a permit space."

In calculating an IDLH for sulfur mustard, data from human acute toxicity studies can be utilized. As previously shown in Table 3, ocular effects, including severe and permanent damage, can occur at sulfur mustard concentrations lower than those producing similar degrees of injury to the respiratory tract. Therefore, ocular effects are the most appropriate to use in setting an IDLH. Dose-response data for ocular effects were previously summarized in Tables 21 and 22. Because an IDLH is intended to be used for a potential 30-min exposure, data points on Table 22 closest to a 30-min exposure were used to evaluate a possible IDLH; these are presented in Table 23.

As shown in Table 23, sulfur mustard concentrations as high as 1.7 mg/m^3 for 33 min ($\text{Ct} = 56 \text{ mg-min/m}^3$) resulted in redness in the eye and conjunctivitis, conditions which are not life threatening and which are not likely to impede escape in the event of respirator failure. Furthermore, these effects are not likely to appear immediately, but only after a latency period of several hours. As discussed in Section 2.3.4 and summarized in Table 3, Cts near or below 12 mg-min/m^3 (after a latency period lasting from several hours to several days) cause only mild eye irritation and redness; this is equivalent to a 30-min exposure to 0.4 mg/m^3 . At Cts of $50\text{-}100 \text{ mg-min/m}^3$, conjunctivitis, tearing, sensitivity to light, and a sensation of grittiness under the eyelids may occur (after a latency period of 4-12 hr); this is equivalent to a 30-min exposure to $1.6\text{-}3.3 \text{ mg/m}^3$. At Cts higher than 100 mg-min/m^3 , corneal edema and clouding, eyelid edema, photophobia, and severe blepharospasm may occur. According to Anderson (1942), the danger zone for exposure to sulfur mustard is $70\text{-}100 \text{ mg-min/m}^3$ in temperate climates and $60\text{-}90 \text{ mg-min/m}^3$ in the tropics. A Ct of 60 mg-min/m^3 is equivalent to a 30-min exposure to 2 mg/m^3 . The ICt_{50} for such effects is 200 mg-min/m^3 (6.6 mg/m^3 for 30 min). Exposure to Cts of $400\text{-}800 \text{ mg-min/m}^3$ are likely to result in corneal damage and possible ulceration after a latency period of 1-4 hr (see also Geeraets et al., 1977). A Ct of 400 mg-min/m^3 is equivalent to a 30-min exposure to 13.3 mg/m^3 . Corneal damage and ulceration are considered to be effects falling under the definition of an IDLH, especially because there is evidence that recurrent keratitis may occur many years after such an exposure (see Section 2.3.7 for discussion). The threshold for such severe effects may fall somewhere below a Ct of 400 mg-min/m^3 for some individuals.

Table 23. Data Points for Ocular Effects Used to Evaluate an IDLH for Sulfur Mustard				
Conc. (mg/m ³)	Expos. Time (min)	Total Number Tested	Effects (Number of individuals affected)	Ref. ^d
0.06 ^a	30	5	Slight injection (1/5) Marked injection (1/5)	A
0.18 ^a	30	1	None	A
0.3 ^a	30	8	Conjunctivitis (1/8) Marked conjunctivitis (1/8) Severe conjunctivitis (1/8)	A
1.4 ^b	30	4	Generalized effect (4/4) ^c	B
1.7 ^b	33	3	Band of conjunctival injection, exposed sclera (3/3)	C

SOURCE: See Reference list below

^a Estimated concentration; reported by Reed et al. (1918) to be 60-70% of nominal value; the use of 60% of the nominal value is considered a protective approach. Sulfur mustard was sprayed as an aerosol mixture of agent and "absolute alcohol"

^b Analytical measurement

^c Number of subjects affected not clearly identified

^d References: A=Reed, 1918; B=Guild et al., 1941; C=Anderson, 1942

A protective approach in setting an IDLH for sulfur mustard would be to use experimental human exposure data, for the appropriate time period, at which life threatening or escape-impairing effects are not expected to occur. The data in Table 23 indicate that a 33-min exposure to 1.7 mg/m³ would fulfill these conditions. This value is below the threshold Ct of 100 mg-min/m³ (30-min exposure to 3.3 mg/m³) and is near the lower limit of the exposure range that Anderson (1942) identified as the danger zone for high humidity and high temperature conditions. The use of this value would also be protective of workers who may have been previously exposed to sulfur mustard at the WPL or at the STEL and who, as a result, may have an increased sensitivity to the agent. The concentration of 1.7 mg/m³ can therefore be used as a LOAEL in the following equation:

$$IDLH = LOAEL_{\text{inhal}} \times \frac{1}{UF_s \times MF}$$

As discussed in Section 3.5.2.1, the overall exposure-response data for ocular effects in humans support a linear extrapolation over the time periods of concern for the IDLH (i.e., from a 33-min

experimental exposure to a 30- min IDLH exposure). Therefore, the IDLH can be estimated as:

$$\text{IDLH} = 1.7 \text{ mg/m}^3 \times \frac{33 \text{ min}}{30 \text{ min}} \times \frac{1}{\text{UFs} \times \text{MF}}$$

$$\boxed{\text{IDLH} = 1.9 \text{ mg/m}^3 = 2.0 \text{ mg/m}^3}$$

A total uncertainty factor of 1 was applied:

- UF_H - Protection of sensitive subpopulations is not considered necessary for a healthy worker population.
- UF_A - Application of this UF is not needed because human data are used.
- UF_S - A UF_S of 1 is used because the IDLH is intended to be for a single exposure
- UF_L - A UF_L of 1 is used because adverse effects can be seen at the IDLH so long as they are not irreversible, and a NOAEL is not required for an IDLH determination
- UF_D - The data base for sulfur mustard is considered to be adequate (see discussion in Section 3.5.1.1).
- MF - A Modifying Factor of 1 is used because no other uncertainties exist in the data.

4. DISCUSSION AND CONCLUSIONS

4.1 AELs for Chronic Exposures

4.1.1 General Population AEL for Chronic Exposures (GPL)

The AEL for the general population (GPL) was calculated using both short-term human and long-term animal data (see Section 3.5.1.1). The GPL derived from the human data is 0.00002 mg/m³, and that derived from the long-term animal study is also 0.00002 mg/m³. The human data point selected involved a continuous exposure to sulfur mustard for a maximum time period of 600 min. Use of such short-term data requires the assumption of a linear response pattern extending from an acute to chronic exposure. Although a linear response pattern appears to be the case for short-term exposures (see Section 3.5.2.1), the experimental data, as shown in Table 21, indicate that this may not be the case for extended exposures to low concentrations (i.e., 0.1 mg/m³), and it may not apply to HD concentrations below the known threshold for adverse effects. Therefore, the GPL derived from the human data is considered to be a protective estimate. The fact that the GPL derived from the animal data does not differ from that derived from the human data lends support to the conclusion that the calculated GPL of 0.00002 mg/m³ is reasonable and protective.

The proposed GPL for sulfur mustard derived in this analysis (0.00002 mg/m³) is lower by a factor of five than the current GPL of 0.0001 mg/m³ established by CDC (DHHS, 1988).

4.1.1.1 Carcinogenicity Assessment

Available human data suggest that human cancers may occur only after exposures sufficiently large to cause acute injuries or following occupational (including battlefield) exposures to elevated concentrations (see Section 2.3). However, as some carcinogens, including sulfur mustard, are currently assumed to be "non-threshold" agents (i.e., linear mode of action, see Section 3.3.2), calculational approaches have been developed to maximally estimate individual increased lifetime cancer risk, "R" (USEPA, 1991), even for extremely limited exposures. The procedure involves estimating a chemical's "Unit Risk" and then multiplying the Unit Risk by the air concentration of concern, with appropriate adjustments for exposure duration. The calculated risk estimate "R" can then be assessed against risk management goals. This process has been performed for the GPL (0.00002 mg/m³) calculated previously in this document. Because there are limited data from which to estimate the Unit Risk, a range of Unit Risk estimates has been included in this evaluation: the value of 0.085 per µg/m³ proposed by USEPA (1991), the geometric mean (0.0041 per µg/m³) of all known estimates summarized in Table 20 of this report, and the estimate derived by Rosenblatt (1987) and USEPA (1991) (0.00046 per µg/m³) from the Japanese war gas factory worker data of Wada et al (1968) and Yamada et al (1957). These Unit Risk estimates were each multiplied by the concentration represented by the pre-determined GPL estimate of 0.00002 mg/m³ and adjusted for standard (conservative) EPA default assumptions regarding lifetime exposure frequency and duration for the general residential scenario. The resulting estimates of risk are presented in Table 24. While the authors prefer the estimate derived from the geometric mean of all estimates (0.0041 per µg/m³) as a more solidly supported value, the range demonstrates the considerable amount of uncertainty in this determination.

As noted in Section 3.2.2, the U.S. Environmental Protection Agency has stated that "for known or suspected carcinogens, acceptable exposure levels are generally concentration levels that represent an excess upper bound lifetime cancer risk to an individual of between 10⁻⁴ and 10⁻⁶." In a review of governmental decisions concerning hazardous waste sites, Travis et al. (1987) found that 10⁻⁴ (1 in 10,000) was used as a *de minimis* risk level; a *de minimis* risk being an acceptable level that is below regulatory concern. The estimated individual cancer risk associated with a sulfur mustard GPL of 2 x 10⁻⁵ mg/m³ and the Unit Risk of 0.0041 per µg/m³ would fall below this *de minimis* level (see Table 24).

Under the exposure assumptions provided in the Table 24, the air concentrations (C) corresponding to given levels of excess lifetime risk may be estimated using the following expression:

$$C = \frac{\text{Risk Level}}{\text{Unit Risk}}$$

For the unit risk of 0.0041 per µg/m³ (or 4.1 per mg/m³), and a lifetime excess cancer risk of 1 x 10⁻⁴, the resulting estimate of the sulfur mustard concentration is no less than:

$$C = \frac{1 \times 10^{-4}}{4.1(\text{mg}/\text{m}^3)^{-1}} = 2.5 \times 10^{-5} \text{ mg HD}/\text{m}^3$$

In a corresponding manner, concentrations can be calculated for other risk levels. For an excess lifetime cancer risk level of 1×10^{-5} , the concentration would be no less than 2.5×10^{-6} mg HD/m³; for a risk of 1×10^{-6} , the corresponding concentration would be no less than 2.5×10^{-7} mg HD/m³.

In summary, the (upper bound) estimates of increased individual lifetime cancer risk associated with potential chronic exposures to levels equivalent to the GPL recommended in this report range from 7.0×10^{-4} to 3.8×10^{-6} . This is considered within the range described by the EPA and other agencies as "acceptable" risk. These estimates do not take into account population size.

Table 24. Upper Bounds on Estimated Individual Increased Cancer Risk at Recommended GPL and WPL Concentrations of Sulfur Mustard Vapor Under Varying Assumptions of Exposure Duration and Unit Risk

HD Exposure Limit (Exposure Assumptions)	Risk Associated with Unit Risk Estimate of		
	0.085 ^a per $\mu\text{g}/\text{m}^3$	0.0041 ^b per $\mu\text{g}/\text{m}^3$	0.00046 ^c per $\mu\text{g}/\text{m}^3$
GPL of 2×10^{-5} mg/m ³ (residential; 24 hr/day, 350 days/yr for 30 yr)	7.0×10^{-4}	3.4×10^{-5}	3.8×10^{-6}
WPL of 4×10^{-4} mg/m ³ [occupational: 8 (and 12) hr/day, 250 days/yr for 25 yr]	2.8×10^{-3} (4.2×10^{-3}) ^d	1.3×10^{-4} (2.0×10^{-4})^d	1.5×10^{-5} (2.3×10^{-5}) ^d
WPL of 4×10^{-4} mg/m ³ [occupational; 8 (and 12) hr/day, 250 days/yr for 5 yr]	5.5×10^{-4} (8.3×10^{-4}) ^d	2.7×10^{-5} (4.0×10^{-5})^d	3.0×10^{-6} (4.5×10^{-6}) ^d

^a Unit risk estimate as proposed by USEPA (1991)

^b The geometric mean of all calculated estimates of unit risk summarized in Table 20 of the current evaluation, and considered by the authors of this report to be the more solidly supported value

^c Derived from Rosenblatt (1987) and USEPA (1991) from non-quantitative air concentration estimate in Japanese war gas factory atmosphere and available cancer incidence data for former factory workers (Wada et al 1968; Yamada et al 1957), and oral to inhalation extrapolation ($0.286 \text{ mg}/\text{kg}/\text{day}$ per mg/m^3)

^d Values in parenthesis for 12 hr/da continuous exposure for same number of days per year and total years

4.1.2 Worker AEL for Chronic Exposures (WPL)

As in the case of the GPL, the WPL for sulfur mustard was calculated using both short-term human exposure data and long-term animal data. The short-term human study involved three 8-hr exposures, one on each of three consecutive days. The effects seen under these test conditions were very mild symptoms of ocular toxicity. Since this exposure frequency is similar to that which workers would experience, the data would be appropriate for calculating a 8-hr/day, 5 days/wk exposure limit. Although the same uncertainties exist in interpreting the results of this exposure in terms of possible cumulative effects following long-term exposures, Papirmeister et al. (1991) has stated that cumulative effects are less likely if the exposures are separated by a 2-3 day exposure-free period. Since workers would experience such a recovery period during weekends, the potential for cumulative effects may be greatly diminished. The WPL derived from the human data is 0.0004 mg/m³, while that from the long-term animal study of McNamara et al. (1975) is 0.0003 mg/m³, although differing by a factor of 0.75, these are essentially the same value. A WPL of 0.0004 mg/m³ is the value recommended because it is based on human data.

The recommended WPL of 0.0004 mg/m³ derived from the human data is about an order of magnitude lower than the current WPL of 0.003 mg/m³ (DHHS 1988).

4.1.2.1 Carcinogenicity Assessment

As with the GPL (see Section 4.1.1.1), the WPLs derived from non-cancer effects in this report have also been evaluated against cancer Unit Risk estimates for HD, even though available human data indicate that human cancers occur only after exposures sufficiently large to cause acute injuries or following occupational (including battlefield) exposures to elevated concentrations (see Section 2.3). The exposure assumptions and resulting estimates of risk are also presented in Table 24.

As noted in Section 3.2.2, the Occupational Safety and Health Administration (OSHA) generally considers 10⁻³ (1 in 1,000) a threshold of significant risk for occupational exposures to a carcinogen (Rodricks et al., 1987; Graham, 1993), and the agency usually does not regulate lower risks because of feasibility limitations (Lohner, 1997). There are specific cases where the occupational exposure limits for some industrial carcinogens correspond to levels of risk higher than 1 per thousand (see Section 3.2.2). For the Unit Risk of 0.0041 per mg/m³, the calculated cancer risk associated with the two WPL scenarios (8-hr and 12-hr shifts) listed in Table 24 is less than 1 per thousand.

In summary, the (upper bound) estimates of increased individual lifetime cancer risk associated with potential chronic exposures to levels equivalent to the WPL recommended in this report range from 4.2 x 10⁻³ to 3.0 x 10⁻⁶. This is considered within the range described by occupational industry and OSHA as "acceptable" occupational risk. These estimates do not take into account population size.

4.2 AELs for Short-term Exposures to Workers

4.2.1 STEL

Human exposure data exist for calculating a STEL for sulfur mustard. A STEL (maximum of four 15-min exposures per day) calculated from the experimental data for single exposures resulted in values which, when averaged over an 8-hr work day, exceeded the 8-hr WPL. Another calculational approach, using a time-adjusted LOAEL, resulted in a value (0.0036 mg/m^3) very similar to the values calculated using probit analysis and logistics analysis (0.003 and 0.0067 mg/m^3 , respectively; with appropriate Uncertainty Factors). This comparison provides a degree of confidence that a STEL of 0.003 mg/m^3 is reasonable and protective. Further, the value of 0.003 mg/m^3 is a factor of 3 below the estimated no-effect concentration of 0.01 mg HD/m^3 for ocular effects. Therefore, for both technical and operational reasons, the recommended STEL for sulfur mustard agent is 0.003 mg/m^3 .

This evaluation recommends that the STEL be used as an alarm level in occupational settings.

4.2.2 IDLH

Previously, there had not been an estimate of IDLH specific for sulfur mustard agents. Army regulatory guidance for sulfur mustard IDLH was that "since workers are required to wear supplied air or SCBA (self-contained breathing apparatus) at vesicant levels much lower than IDLH levels," the establishment of IDLH values for the vesicant sulfur mustard (and Lewisite) was unnecessary (AR 385-61; DA 1997a). At present, a pressure-demand, full-face piece, SCBA or supplied-air respirator is to be worn by any sulfur mustard agent worker conducting operations in areas where concentrations exceed 0.003 mg/m^3 (AR 385-61; DA 1997a).

Adequate human exposure data exist for calculating an IDLH for sulfur mustard. The data include exposure times of 30-33 min (see Table 23). At exposure concentrations of 0.06 to 1.7 mg/m^3 , the observed effects were no more severe than severe conjunctivitis. The highest value of 1.7 mg/m^3 was used to calculate an IDLH of 2.0 mg/m^3 . Although the data suggest that 30-min exposures to sulfur mustard air concentrations even higher than 2.0 mg/m^3 may be below a true IDLH condition, the choice of 2.0 mg/m^3 is considered to be appropriate in light of the possibility of increased sensitivity of workers who may have had previous exposures to the agent. Furthermore, because the data indicate the dose-response curve for sulfur mustard is relatively steep, i.e., 30-min exposures to 13.3 mg/m^3 may cause severe eye damage and re-occurring keratitis years after the exposure, an IDLH of 2.0 mg/m^3 would provide a greater margin of safety.

5. RECOMMENDATIONS

Based on the discussions and conclusions given in Section 4, this report's recommendations for sulfur mustard air exposure limits are as shown in Table 25. As noted previously, the human exposure data were considered to be more appropriate than the animal data for establishing exposure limits for the general population (GPL), as well as for workers (WPL)

Table 25. Existing and Recommended Airborne Exposure Limits for Sulfur Mustard					
Application	Type	<u>Existing</u> (mg/m³)	<u>Recommended</u> (mg/m³)	Exposure Time	Frequency
General population	GPL ^a (TWA) ^b	0.0001	0.00002	24 hr/day	7 days/wk, lifetime
Occupational	WPL ^c (TWA)	0.003	0.0004	8 hr/day	5 days/wk
	STEL ^d	NA	0.003	15 min	4 times/day
	IDLH ^e	NA	2.0	30 min	one time

^a GPL. = General population AEL (no observable adverse effects)

^b TWA = Time-weighted-average

^c WPL = Occupational AEL (no observable adverse effects)

^d STEL = Short-term Exposure Limit

^e IDLH = Immediately Dangerous to Life or Health

LITERATURE CITED

- American Conference of Governmental Industrial Hygienists (ACGIH). 2000. *2000 TLVs and BEIs: Threshold Limit Values for Chemical Substances and Physical Agents and Biological Exposure Indices*. ACGIH, 1330 Kemper Drive, Cincinnati, OH 45240-1634.
- Anderson, J.S. 1942. *The Effect of Mustard Gas Vapour on Eyes under Indian Hot Weather Conditions*. CDRE Report No. 241. Chemical Defense Research Establishment (India).
- Anslow, W.P. and C.R. Houck. 1946. Systematic pharmacology and pathology of sulfur and nitrogen mustards. In: *Chemical Warfare Agents, and Related Chemical Problems*, vol. 1, pp. 440-478I. Summary Technical Report of Division 9. National Defense Research Committee, U.S. Office of Scientific Research and Development, Washington, DC.
- ATSDR (Agency for Toxic Substances and Disease Registry). 1992. *Toxicological Profile for Mustard Gas*. TP-91/22. Agency for Toxic Substances and Disease Registry, Atlanta, GA.
- Auerbach, C. and J.M. Robson. 1946. Chemical production of mutagens. *Nature (Lond.)* 157:302.
- Axelrod, D.J. and J.G. Hamilton. 1947. Radio-autographic studies of the distribution of lewisite and mustard gas in skin and eye tissues. *Am. J. Pathol.* 23:389-411.
- Azizi, F., A. Keshavarz, F. Roshanzamir, et al. 1995. Reproductive function in men following exposure to chemical warfare with sulfur mustard. *Med. War* 11:34-44.
- Bakshi, K.F. S.N.J. Pang, and R. Snyder. 2000. Review of the U.S. Army's health risk assessments for oral exposure to six chemical-warfare agents. *J. Toxicol. Environ. Health (Part A)* 59(5-6):281-526.
- Balaili, M. 1986. First report of delayed toxic effects of yperite poisoning in Iranian fighters. In: *Terrorism: Analysis and Detection of Explosives*. Proc. Second World Conf. on New Compounds in Biol. and Chem. Warfare, B. Heyndricks, ed., pp. 489-495, Rijksuniversiteit, Ghent.
- Barnes, D.G. and M. Dourson. 1988. Reference dose (RfD): description and use in health risk assessments. *Reg. Toxicol. Pharmacol.* 8:471-486.
- Barkley, J.J. Jr. 1999. *Information for Combat Developers on Performance Effects from Exposure to Chemical Warfare Agents*. U.S. Army Center for Health Promotion and Preventive Medicine, ATTN: MCHB-TS (S. Kistner), 5151 Blackhawk Road, Aberdeen Proving Ground, MD 21010-5403
- Battista, S. P. and E. S. McSweeney. 1965. Approaches to a quantitative method for testing eye irritation. *J. Soc. Cosmet. Chem.* 16:199-301. (As cited in Chan and Hayes, 1985)
- Beckley, J. H. 1965. Comparative eye testing: man vs. animal. *Toxicol. Appl. Pharmacol.* 7:93-101.
- Beebe, G.W. 1960. Lung cancer in World War I veterans: possible relation to mustard gas injury and 1918 influenza epidemic. *J. Natl. Cancer Inst.* 25:1231-1252.

- Boursnell, J.C., J.A. Cohen, M. Dixen, et al. 1946. Studies on mustard gas ($\beta\beta'$ -dichlorodiethyl sulphide) and some related compounds. 5. The fate of injected mustard gas (containing radioactive sulphur) in the animal body. *Biochem. J.* 40:756-764.
- Brown, E.C. 1949. Pulmonary Effects following Chronic Exposure to HS Vapor. Medical Division Report No. 187. Army Chemical Center, MD.
- Buehler, E. V. and E. A. Newman. 1964. A comparison of eye irritation in monkeys and rabbits. *Toxicol. Appl. Pharmacol.* 6:701-710.
- Cameron, G.R., J.H. Gaddum and R.H.D. Short. 1946. The absorption of war gases by the nose. *J. Pathol. Bact.* 58:449-455.
- Capizzi, R.L., W.J. Smith, R. Field and B. Papirmeister. 1973. A host-mediated assay for chemical mutagens using L5178Y/Asn murine leukemia. *Mutat. Res.* 21:6.
- Case, R.A.M. and A.J. Lea. 1955. Mustard gas poisoning, chronic bronchitis and lung cancer. An investigation into the possibility that poisoning by mustard gas in the 1914-1918 war might be a factor in the production of neoplasia. *Brit. J. Prev. Med.* 9:62-72.
- Chan, P-K. and A.W. Hayes. 1985. Assessment of chemically induced ocular toxicity: a survey of methods. In: *Toxicology of the Eye, Ear, and Other Special Senses*. A.W. Hayes, ed., pp. 103-143, Raven press, New York.
- Cicmanec, J.L., M.L. Dourson and R.C. Hertzberg. 1996. Noncancer risk assessment: present and emerging issues. In: *Toxicology and Risk Assessment: Principles, Methods, and Applications*. A.M. Fan and L.W. Chang, eds., pp. 293-310, Marcel Dekker, Inc, NY.
- Clemenson, C.-J., H. Kristoffersson, B. Sorbo and S. Ullberg. 1963. Whole body autoradiographic studies of the distribution of sulphur 35-labelled mustard gas in mice. *Acta Radiol. Ther.* 1:314-320.
- Crathorn, A.R. and J.J. Roberts. 1965. Reactions of cultured mammalian cells of varying radiosensitivity with the radiomimetic alkylating agent mustard gas. *Prog. Biochem. Pharmacol.* 1:320-326.
- Crathorn, A.R. and J.J. Roberts. 1966. Mechanism of the cytotoxic action of alkylating agents in mammalian cells and evidence for the removal of alkylated groups from deoxyribonucleic acid. *Nature (Lond)* 211:150-153.
- Culp, S.J., D.W. Gaylor, W.G. Sheldon, L.S. Goldstein and F.A. Beland. 1998. A comparison of the tumors induced by coal tar and benzo[a]pyrene in a 2-year bioassay. *Carcinogenesis* 19:117-124.
- DA (U.S. Department of the Army). 1974. *Chemical Agent Data Sheets*, vol. 1. Edgewood Arsenal Special Report, EO-SR 74001. Defense Technical Information Center, Alexandria, VA.
- DA (U.S. Department of the Army). 1987. *Chemical Stockpile Disposal Program: Evaluation of Multiple Incinerator Air Quality Impacts Edgewood Area*. SAPEO-CDEIS-87004. Program Manager for Chemical Demilitarization, Aberdeen Proving Ground, MD. (As cited in USEPA, 1991)

DA (U.S. Department of the Army). 1990. *Potential Military Chemical/Biological Agents and Compounds*, Field Manual 3-9 (FM 3-9, NAVFAC P-467, AFR 355-7), Headquarters, Department of the Army, Department of the Navy, Department of the Air Force, Washington, DC (12 December, 1990).

DA (U.S. Department of the Army). 1991. *Occupational Health Guidelines for the Evaluation and Control of Occupational Exposure to Mustard Agents H, HD, and HT*, Pamphlet 40-173, Headquarters, Department of the Army, Washington, DC.

DA (U.S. Department of the Army). 1997a. *The Army Chemical Agent Safety Program*. Army Regulation 385-61. Headquarters, Department of the Army, Washington, DC, Feb. 28, 1997.

DA (U.S. Department of the Army). 1997b. *Toxic Chemical Agent Safety Standards*. Pamphlet 385-61. Headquarters, Department of the Army, Washington, DC, March 31, 1997.

DA/DAF (U.S. Department of the Army and U.S. Air Force). 1975. *Military Chemistry and Chemical Compounds. Field Manual No. FM 3-9*. (As cited in Papirmeister et al., 1991)

Dahl, H., B. Glund, P. Vangstad and M. Norn. 1985. Eye lesions induced by mustard gas. *Acta Ophthalmol.* 63 (suppl. 173):30-31.

Davison, C., R.S. Rozman and P.K. Smith. 1961. Metabolism of bis- β -chloroethyl sulfide (sulfur mustard gas). *Biochem. Pharmacol.* 7:65-74.

DHHS (U.S. Department of Health and Human Services). 1988. Final recommendations for protecting the health and safety against potential adverse effects of long-term exposure to low doses of agents: GA, GB, VX, mustard agent (H, HD, T), and lewisite (L). U.S. Department of Health and Human Services, Centers for Disease Control. *Fed. Reg.* 53(50):8504-8507.

DoD (Department of Defense). 1984. *Ammunition and Explosive Safety Standards*, DoD 6055.9-STD, Assistant Secretary of Defense (manpower, installation, and Logistics), Washington, DC.

Dourson, M.L. S.P. Felter, and D. Robinson. 1996. Evolution of science-based uncertainty factors in noncancer risk assessment. *Reg. Toxicol. Pharmacol.* 24:108-120.

Dowlati, A., G.E. Pierard and Y. Dowlati. 1993. Epidermal hyperplasia with or without atypia in patients exposed to mustard gas. *Arch. Dermatol.* 129:242.

Easton, D.F., J. Peto and R. Doll. 1988. Cancers of the respiratory tract in mustard gas workers. *Br. J. Ind. Med.* 45(10):652-659.

Emison, E.S. and W.J. Smith. 1996. Cytometric analysis of DNA damage in cultured human epithelial cells after exposure to sulfur mustard. *J. Amer. Coll. Toxicol.* 15 (Suppl. 2): S9-S18.

Fox, M. and D. Scott. 1980. The genetic toxicology of nitrogen and sulfur mustard. *Mutat. Res.* 75:131-168.

Friedenwald, J.S. and W. Buschke. 1948. V. Nuclear fragmentation produced by mustard and nitrogen mustards in corneal epithelium. *Bull. Johns Hopkins Hosp.* 82:161-177.

- Friedenwald, J.S., R.O. Scholz, A. Snell, Jr. and S.G. Moses. 1948. Primary reaction of mustard with corneal epithelium. *Bull. Johns Hopkins Hosp.* 82:102-120.
- Ganas, P. 1969. New developments in chemical and biological warfare. *Forces Aeriennes Francaises* 24(263):449-475. (As cited in Papirmeister et al., 1991)
- Gates, M. And S. Moore. 1946. Mustard gas and other sulfur mustards. In: *Chemical Warfare Agents, and Related Chemical Problems*, vol. 1, pp. 30-58. Summary Technical Report of Division 9, National Defense Research Committee, U.S. Office of Scientific Research and Development, Washington, DC.
- Gaylor, D. 1998. *Carcinogenic Potency for Sulfur Mustard*. Memo dated March 11, 1998, to V. Hauschild, U.S. Army Center for Health Promotion and Preventive Medicine. National Center for Toxicological Research, Jefferson, AK.
- Gaylor, D. 2000. Dose Response and Other Current issues in Carcinogen Quantitative Risk Assessment. *Health and Environment Science News Letter*, Number 12, Fall, 2000.
- Gaylor, D. and L.S. Gold. 1995. Quick estimate of the regulatory virtually safe dose based on the maximum tolerated dose for rodent bioassays. *Regul. Toxicol. Pharmacol.* 22:57-63.
- Geeraets, W.J., S. Abedi and R.V. Blanke. 1977. Acute corneal injury by mustard gas. *South. Med. J.* 70(3):348-350. (As cited in Papirmeister et al., 1991)
- Gilman, M. R. 1982. Skin and eye testing in animals. In: *Principles and Methods of Toxicology*. A.W. Hayes, ed., pp. 209-222, Raven Press, New York.
- Graham, J.D. 1993. The legacy of one in a million. *Risk in Perspective* 1:1-2.
- Grant, W. M. 1974. *The Toxicology of the Eye*. 2nd ed., Charles C. Thomas, Springfield, IL
- Grant, W. M. 1986. *The Toxicology of the Eye*. 3rd ed., Charles C. Thomas, Springfield, IL
- Guild, W.J.F., K.P. Harrison, A.Fairley and A.E. Childs. 1941. *The Effect of Sulfur Mustard on the Eyes*. Porton Report 2297. Chemical Defense Experimental Station, Porton, UK.
- Hackett, P.L., R.L. Rommereim, F.G. Burton, R.L. Buschbom and L.B. Sasser. 1987. *Teratology Studies on Lewisite and Sulfur Mustard Agents: Effects of Sulfur Mustard in Rats and Rabbits*. Final Report. AD A187495. Pacific Northwest Laboratory, Richland, WA. Prepared for the U.S. Army Medical Research and Development Command, Fort Detrick, MD.
- Hambrook, J.L., D.J. Howells and C. Schock. 1993. Biological fate of sulphur mustard (1,1'-thiobis(2-chloroethane)): uptake, distribution and retention of ³⁵S in skin and in blood after cutaneous application of ³⁵S-sulphur mustard in rat and comparison with human blood *in vitro*. *Xenobiotica* 23:537-561.
- Henry, M.C. 1991. *Literature Review of Sulfur Mustard Toxicity*. USAMRICD-TR-91-01. U.S. Army Medical Research Institute of Chemical Defense, Aberdeen Proving Ground, MD.
- Heston, W.E. 1950. Carcinogenic action of mustards. *J. Natl. Cancer Inst.* 11:415-423.

- Heston, W.E. 1953. Occurrence of tumors in mice injected subcutaneously with sulfur mustard and nitrogen mustard. *J. Natl. Cancer Inst.* 14:131-140.
- Heston, W.E. and W.D. Levillain. 1953. Pulmonary tumors in strain A mice exposed to mustard gas. *Proc. Soc. Exp. Biol.* 82:457-460.
- Hosseini, K., A. Moradi, A. Mansouri and K. Vessal. 1989. Pulmonary manifestations of mustard gas injury: a review of 61 cases. *Iranian J. Med. Sci.* 14:20-26.
- Howe, R.B. and K.S. Crump. 1983. WEIBULL 82. K.S. Crump and Company, Inc. Ruston, LA.
- Howe, R.B., K.S. Crump and C. Van Landingham. 1986. GLOBAL 86: A computer program to extrapolate quantal animal toxicity to low doses. EPA-68-01-6826. U.S. Environmental Protection Agency, Washington, DC.
- IARC (International Agency for Research on Cancer). 1975. *IARC Monographs on the Evaluation of the Carcinogenic Risk of Chemicals to Man: Some Aziridines, N, S, & O-mustards and Selenium*, vol. 9, pp. 181-207. International Agency for Research on Cancer, Lyons, France.
- IARC (International Agency for Research on Cancer). 1987a. *IARC Monographs on the Evaluation of the Carcinogenic Risk of Chemicals to Man: Genetic and Related Effects: An Updating of Selected IARC Monographs Volumes 1-42, Suppl. 6*, International Agency for Research on Cancer, Lyons, France.
- IARC (International Agency for Research on Cancer). 1987b. *IARC Monographs on the Evaluation of the Carcinogenic Risk of Chemicals to Man: Overall Evaluation of Carcinogenicity: An Updating of IARC Monographs Volumes 1-42, Suppl. 7*, p. 67. International Agency for Research on Cancer, Lyons, France.
- Inada, S., K. Hiragun, K. Seo and T. Yamura. 1978. Multiple Bowen's disease observed in former workers of a poison gas factory in Japan, with special reference to mustard gas exposure. *J. Dermatol.* 5:49-60.
- IOM (Institute of Medicine, Committee to Survey the Health Effects of Mustard Gas and Lewisite, Division of Health Promotion and Disease Prevention). 1993. *Veterans at Risk; The Health Effects of Mustard Gas and Lewisite*, C.M. Pechura and D.P. Rall, eds. National Academy Press, Washington, DC.
- IPCS (International Programme on Chemical Safety). 1994. *Assessing Human Health Risks of Chemical: Derivation of Guidance Values for Health-based Exposure Limits*. Environmental Health Criteria, vol. 170. World Health Organization, Geneva, Switzerland.
- ITII (International Technical Information Institute). 1975. *Toxic and Hazardous Industrial Chemicals Safety Manual*. p. 351. International Technical Information Institute, Tokyo, Japan.
- Jones, TD, P.J Walsh, A.P. Watson, et al. 1988. Chemical scoring by a Rapid Screening Hazard (RASH) method. *Risk Anal.* 8:99-118.
- Jostes, R.F., L.B. Sasser and R.J. Rausch. 1989. *Toxicology Studies on Lewisite and Sulfur Mustard Agents: Genetic Toxicity of Sulfur Mustard (HD) in the Chinese Hamster Ovary Cells*. Final Report,

PNL-6916. Pacific Northwest Laboratories, Richland, WA. Prepared for the U.S. Army Medical Research and Development Command, Fort Detrick, MD.

Karnofsky, D.A. and J.T. Nolen. 1944. *Report on Mustard Vapor Casualties Occurring at Bushnell, Florida*. Dugway Proving Ground, Chemical Warfare Service, Mobile Field Unit, Bushnell, FL., vol. 1. Edgewood Arsenal Special Report, EO-SR 74001. Defense Tech, Inform. Center, Alexandria, VA. (As cited in Papirmeister et al., 1991)

Kircher, M. and M. Brendel. 1983. DNA alkylation by mustard gas in yeast *Saccharomyces cerevisiae* strains of different repair capacity. *Chem.-Biol. Interact.* 44:27-39.

Klehr, N. 1984. Cutaneous late manifestation in former mustard gas workers. *Z. Hautkrankh.* 59:1161-1170. (In German with English abstract)

Landahl, H.D. 1945. *A Formal Analysis of the Action of Liquid Vesicants on Bare Skin*. University of Chicago Toxicity Laboratory, Report No. 50. Chicago, IL.

Laughlin, R.C. 1944a. *Correlation of Eye Changes in Rabbits with CT Exposure to HD*. MRL (EA) Report 23.

Laughlin, R.C. 1944b. *Continued Exposure of Human Eyes to H Vapor, MIT Subjects*. MRL (EA) Report 9.

Laughlin, R.C. 1944c. *Eye Examination of Factory Workers Handling H, CN and CG*. MRL (EA) Report 18.

Lawley, P.D. and P. Brookes. 1965. Molecular mechanism of the cytotoxic action of difunctional alkylating agents and of resistance to this action. *Nature (Lond.)* 206:480-483.

Lehman, H.S. and O.G. Fitzhugh. 1954. 100-Fold margin of safety. *Assoc. Food Drug Off. U.S. Q. Bull.* 18:33-35. (As cited in Mioduszewski et al 1998)

Lewin, B. 1990. *Genes IV*. Oxford University Press, Walton St., Oxford OX2 6DP

Lewis, R.J. and D.V. Sweet (eds.) 1984. *Registry of Toxic Effects of Chemical Substances*. 1983 supplement to the 1981-82 edition. pp. 1153, 1169. U.S. Department of Health and Human Services, National Institute for Occupational Safety and Health, Cincinnati, OH.

Lohner, T.W. 1997. Is 10^{-6} an appropriate *de minimis* cancer risk goal? *Risk Policy Report*, April 18, 1997, pp. 31-33.

MacNaughton, M.G. and J.H. Brewer. 1994. *Environmental Chemistry and Fate of Chemical Warfare Agents*. Southwest Research Institute, San Antonio, TX

Major, M. 2000. Derivation of Inhalation Reference Concentrations for Workplace and General Population Exposures to HD. Presented at the In-Progress Review (IRP) Technical Consensus Meeting, Airborne Exposure Limits for Sulfur Mustard (HD), April, 19, 2000, Aberdeen Proving Ground, MD.

Manning, K.P., D.C.G. Skegg, P.M. Stell, et al. 1981. Cancer of the larynx and other occupational hazards of mustard gas workers. *Clin. Otolaryngol.* 6:165-170.

Marrs, T.C., R.L. Maynard and F.R. Sidell. 1996. *Chemical Warfare Agents, Toxicology and Treatment*. John Wiley and Sons, New York, 243 pp.

Martinez-Lopez, L. 2000. Chronic Toxicological Criteria for Chemical Warfare Compounds. Memorandum through Assistant Surgeon General for Force Projection, Office of the Surgeon General, 800 Army Pentagon, Washington, D.C. from BG L. Martinez-Lopez, Functional Proponent for Preventive Medicine, Department of the Army, Office of the Surgeon General, 5709 Leesburg Pike, Falls Church, VA 22041-3258 (16 Feb 2000).

Marzulli, F. N. and M. E. Simmon. 1971. Eye irritation from topically applied drugs and cosmetics: preclinical studies. *Am. J. Optom.* 48:61-79. (As cited in Chan and Hayes, 1985)

Maurice, D.M. and A. A. Giardini 1951. A simple optical apparatus for measuring the corneal thickness, and the average thickness of the human cornea. *Br. J. Ophthalmol.* 35:169-177.

McColl, R.S. 1990. *Biological Safety Factors in Toxicological Risk Assessment*. SSC H49-49/1990E. Supply and Services Canada, Ottawa. (As cited in Mioduszewski et al., 1998)

McNamara, B.P., E.J. Owens, M.K. Christensen, et al. 1975. *Toxicological Basis for Controlling Levels of Mustard in the Environment*. EASP EBSP 74030. Biomedical Laboratory, Department of the Army, Headquarters, Edgewood Arsenal, Aberdeen Proving Ground, MD.

Medema, J. 1986. Mustard gas: the science of H. *Nuclear, Biological, and Chemical Defense and Technology International* 1:66-71.

Mioduszewski, R.J., Reutter, S.H., Thomson, et al. 1998. *Evaluation of Airborne Exposure Limits for G-Agents: Occupational and General Population Exposure Criteria*. ERDEC-TR-489. U.S. Department of the Army, Edgewood Research, Development and Engineering Center, U.S. Army Chemical and Biological Defense Command, Aberdeen Proving Ground, MD.

Mishima, S. and B. O. Hedbys, 1968. Measurement of corneal thickness with the Haag-Streit pachometer. *Arch. Ophthalmol.* 80:710-713.

Moore, A.M. and J.B. Rockman. 1950. A study of human hypersensitivity to compounds of the mustard gas type. *Can. J. Res.* 28E:169-176.

Morgenstern, P., F.R. Koss, and W.W. Alexander. 1947. Residual mustard gas bronchitis; effects of prolonged exposure to low concentrations. *Ann. Internal Med.* 26:27-40.

NAC/AGEL (National Advisory Committee on Acute Exposure Guideline Levels for Hazardous Substances). 2000. Standing Operating Procedures of the National Advisory Committee on Acute Exposure Guideline Levels for Hazardous Substances. Draft Report, Office of Pollution Prevention and Toxics, U.S. Environmental Protection Agency, Washington, D.C. (30 June 2000).

Nakamura, T. 1956. Studies on the warfare gas-injury in Japan. Report I. On the general condition of the poison gas island. *Hiroshima Med. J.* 4:1141-1149. (In Japanese, as cited in Inada et al., 1978)

NDRC (National Defense Research Committee). 1946. *Chemical Warfare Agents, and Related Chemical Problems*. Summary Technical Report of Division 9 (Parts I-II), Office of Scientific Research and Development, Washington, D.C.

Nishimoto, Y., M. Yamakido, T. Shinegobu, et al. 1983. Long term observations of poison gas workers with special reference to respiratory cancers. *J. UOEH* 5(Suppl.):89-94.

Nishimoto, Y., M. Yamakido, S. Ishioka, et al. 1988. Epidemiological studies of lung cancer in Japanese mustard gas workers. In: *Unusual Occurrences as Clues to Cancer Etiology*, R.W. Miller et al., eds., pp. 95-101. Japan. Sci. Soc. Press., Tokyo.

Norman, J. 1975. Lung cancer mortality in World War I veterans with mustard gas injury: 1919-1945. *J. Natl. Cancer Inst.* 54:311-317.

NRC (National Research Council), 1970. *Evaluating the Safety of Food Chemicals*. Food Protection Committee of the National Research Council, National Academy Press, Washington, D.C. (As cited in Mioduszewski et al 1998)

NRC (National Research Council). 1985. *Possible Long-Term Effects of Short-term Exposures to Chemical Agents*, vol. 3, Current Health Status of Test Subjects. National Research Council, Washington, DC.

NRC (National Research Council). 1993. *Guidelines for Developing Community Emergency Exposure Levels for Hazardous Substances*. Committee on Toxicology, National Research Council, Washington, DC.

NRC (National Research Council). 1997. *Review of Acute Human Toxicity Estimates for Selected Chemical-Warfare Agents*. Committee on Toxicology, National Research Council, Washington, DC.

NRC (National Research Council). 1999. *Review of the U.S. Army's Health Risk Assessments for Oral Exposure to Six Chemical-Warfare Agents*. Committee on Toxicology, National Research Council, Washington, DC.

NTP (National Toxicology Program). 1980. *Annual Report on Carcinogens*. National Toxicology Program. Research Triangle Park, NC.

Omenn, G.S., S. Stuebbe and L.B. Lave. 1995. Predictions of rodent carcinogenicity testing results: interpretation in light of the Lave-Omenn value-of-information model. *Molec. Carcinogen.* 14:37-45.

Opresko, D.M, R.A. Young, R.A. Faust, S.S. Talmage, A.P. Watson, R.H. Ross, K.A. Davidson and J. King. 1998. Chemical warfare agents: estimating oral reference doses. *Rev. Environ. Contam. Toxicol.* 156:1-183.

Otto, C.E. 1946. *A Preliminary report on the Ocular Action of Dichlorethyl Sulfide (Mustard Gas) in Man as seen at Edgewood Arsenal, Edgewood, Maryland*. Chemical Warfare Service, Report No. EAL-539. (As cited in Papirmeister et al., 1991)

Papirmeister, B., A.J. Feister, S.I. Robinson and R.D. Ford. 1991. *Medical Defense Against Mustard Gas: Toxic Mechanisms and Pharmacological Implications*. CRC Press, Boca Raton, FL. 359 pp.

PCS (Project Coordination Staff). 1945. *Resume of Recent Knowledge on the Technical Aspects of Chemical Warfare in the Field* (Appendices to PCS Report No. 9). Chemical Warfare Service, Washington, D.C. (17 May 1945) (as cited in Barkley, 1999)

PCS (Project Coordination Staff). 1946. *Technical Aspects of Chemical Warfare in the Field*. Chemical Warfare Service, Washington, D.C.

Porton Report. 1931a. *Sensitivity to Mustard Gas*. Porton Report #930, WA-1727-13a, Porton Down, UK (22 July 1931).

Porton Report. 1931b. *Further Report on Sensitivity to Mustard Gas*. Porton Report #948, WA-1727-13b, Porton Down, UK (23 Sept 1931).

Reed, C.I. 1918. *The Minimum Concentration of Mustard Gas Effective for Man* (Preliminary Report), Report No. 318. Pharmacology Research Section, American University.

Reed, C.I., E.F. Hopkins and C.F. Weyand. 1918. *The Minimum Concentration of Mustard Gas Effective for Man (Final Report)*. Report No. 329, War Department, Medical Division, C.W.S., Pharmacology Research Section, American University Experiment Station, Washington, D.C.

Reed, C.I. 1920. The Minimum Concentration of Dichlorethylsulfide (Mustard Gas) Effective for Man. *J. Pharm. Exp. Therap.* 15:77-80.

Renwick, A.G. 1993. Data derived safety factors for the evaluation of food additives and environmental contaminants. *Food Add. Contam.* 10:275-305.

Renwick, A.G. and N.R. Lazarus. 1998. Human variability and noncancer risk assessment - an analysis of the default uncertainty factor. *Regul. Toxicol. Pharmacol.* 27:3-20.

Renshaw, B. 1946. Mechanisms in production of cutaneous injuries by sulfur and nitrogen mustards. In: *Chemical Warfare Agents and Related Chemical Problems*, vol. 1, chapter 23, pp. 479-518. Summary Technical Report of Division 9, National Defense Research Committee, U.S. Office of Scientific Research and Development, Washington, D.C.

Reutter, S.A., Mioduszewski, R.J., Thomson, S.A. 2000. *Evaluation of Airborne Exposure Limits for VX: Worker and General Population Exposure Criteria*. ECBC-TR-074. Edgewood Chemical Biological Center, U.S. Army Soldier and Biological Chemical Command, Aberdeen Proving Ground, MD.

- Riviere, J.E., J.D. Brooks, P.L. Williams and N.A. Monteiro-Riviere. 1995. Toxiokinetics of topical sulfur mustard penetration, disposition, and vascular toxicity in isolated perfused porcine skin. *Toxicol. Appl. Pharmacol.* 135:25-34.
- Roberts, J.J. and G.P. Warwick. 1963. Studies of the mode of the action of alkylating agents - VI. The metabolism of bis- β -2-chloroethylsulphide (mustard gas) and related compounds. *Biochem. Pharmacol.* 12:1329-1334.
- Robinson, J. P. 1967. Chemical warfare. *Science Journal* 4:33-40. (As cited in IOM, 1993).
- Rodricks, J.V., S.M. Brett and G.C. Wrenn. 1987. Significant risk decisions in federal regulatory agencies. *Reg. Toxicol. Pharmacol.* 7:307-320.
- Rosenblatt, D.H., T.A. Miller, J.C. Dacre, I. Muul and D.R. Cogley. 1975. *Problem Definition Studies on Potential Environmental Pollutants. II. Physical, Chemical, Toxicological, and Biological Properties of 16 Substances.* Tech. Report 7509, AD AO30428. U.S. Army Medical Bioengineering Research and Development Laboratory, Fort Detrick, MD.
- Rosenblatt, D.H. 1987. *Recalculation of general population exposure limits and stack detection levels for mustard* (Unpublished). U.S. Army Biomedical Research and Development Laboratory, Fort Detrick, Frederick, MD 21701-5010.
- Rosenblatt, D.H., M.J. Small, T.A. Kimmell and A.W. Anderson. 1995. *Agent Decontamination Chemistry Technical Report.* U.S. Army Test and Evaluation Command (TECOM) Technical Report, Phase I. Draft Report, Argonne National Laboratory.
- Rozmiarek, H., R.L. Capizzi, B. Papirmeister et al. 1973. Mutagenic activity in somatic and germ cells following chronic inhalation of sulfur mustard. *Mutat. Res.* 21:13-14.
- Sasser L.B., R. A. Miller, D.R. Kalkwarf, et al. 1989a. *Toxicology Studies on Lewisite and Sulfur Mustard Agents: Subchronic Toxicity of Sulfur Mustard (HD) in Rats.* Final Report , PNL-6870, Pacific Northwest Laboratories, Richland, WA. Prepared for the U.S. Army Medical Research and Development Command, Fort Detrick, MD.
- Sasser L.B., R. A. Miller, D.R. Kalkwarf, et al. 1989b. *Toxicology Studies on Lewisite and Sulfur Mustard Agents: Two-Generation Reproduction Study of Sulfur Mustard (HD) in Rats.* Final Report, PNL-6944, Pacific Northwest Laboratories, Richland, WA. Prepared for the U.S. Army Medical Research and Development Command, Fort Detrick, MD.
- Sasser L.B., R. A. Miller, J.A. Cushing and J.C. Dacre. 1990. Dominant lethal effect of sulfur mustard in rats. *Toxicologist* 10:225. (Abstract)
- Sasser L.B., R.A. Miller, D.R. Kalkwarf, J.A. Cushing, and J.A. Dacre, 1996. Subchronic toxicity evaluation of sulfur mustard in rats. *J. of Appl. Toxicol.* 16: 5-13.
- Scott, D., M. Fox and B.W. Fox. 1974. The relationship between chromosomal aberrations, survival and DNA repair in tumor cell lines of differential sensitivity to x-rays and sulphur mustard. *Mutat. Res.* 22:207-221.

- Shakil, F.A., A. Kuramoto, M. Yamakido, et al. 1993. Cytogenetic abnormalities of hematopoietic tissue in retired workers of the Ohkunojima poison gas factory. *Hiroshima J. Med. Sci.* 42:159-165.
- Shimkin, M.B. and J.N. McClelland. 1949. Induced pulmonary tumors in mice. IV. Analysis of dose-response data with methylcholanthrene. *J. Natl. Cancer Inst.* 10:597-603.
- Sidell, F.R. 1990. Clinical Notes on Chemical Casualty Care. U.S. Army Medical Research Institute of Chemical Defense (USAMRICD) Technical memorandum 90-1. Aberdeen Proving Ground, MD. (As cited in IOM, 1993)
- Sidell, F.R. and C.G. Hurst. 1992. Clinical Considerations in Mustard Poisoning. In: *Chemical Warfare Agents*. A.M. Somani, ed., pp. 51-67, Academic Press, New York.
- Sim, V.M. 1971. Chemicals used as weapons in war. In: *Drill's Pharmacology of Medicine. 4th ed., McGraw-Hill Book Co., New York.*
- Small, M.J. 1984. *Compounds Formed from the Chemical Decontamination of HD, GB, and VX and Their Environmental Fate*. Technical Report 8304, AD A149515, U.S. Army Medical Bioengineering Research and Development Laboratory, Fort Detrick, Frederick, MD.
- Smith, W.J., C.L. Gross, P. Chan and H.L. Meier. 1990. The use of human epidermal keratinocytes in culture as a model for studying the biochemical mechanisms of sulfur mustard induced vesication. *Cell. Biol. Toxicol.* 6: 285-291. (As cited in Emison and Smith 1996).
- Smith, W.J., K.M. Sanders, S.E. Ruddle and C.L. Gross. 1993. Cytometric analysis of DNA changes induced by sulfur mustard. *J. Toxicol-Cutaneous Ocular. Toxicol.* 12:337-343. (As cited in Emison and Smith 1996).
- Somani, S.M. 1992. Toxicokinetics and toxicodynamics of mustard. In: *Chemical Warfare Agents*. A.M. Somani, ed., pp. 13-50, Academic Press, New York.
- Stepanov, A.A. and V.N. Popov. 1962. *Chemical Weapons and Principles of Antichemical Defense*. Joint Publications Research Service (1965) No. 15107, NTIS, Springfield, VA. (As cited in Papirmeister et al., 1991).
- Stewart, D.L., E.J. Sass, L.K. Fritz and L.B. Sasser. 1989. *Toxicology Studies on Lewisite and Sulfur Mustard Agents: Mutagenicity of Sulfur Mustard in the Salmonella Histidine Reversion Assay*. Final Report, PNL-6873, Pacific Northwest Laboratories, Richland, WA. Prepared for the U.S. Army Medical Research and Development Command, Fort Detrick, MD, AD A213102.
- Stoner, G.D., E.S. Greisiger, H.A. Schuf, et al. 1984. A comparison of the lung adenoma response in strain A/J mice after intraperitoneal and oral administration of carcinogens. *Toxicol. Appl. Pharmacol.* 72:313-323
- Stroykov, K.N. 1970. *Medical Aid for Toxic Agent Victims*. Meditsina, Moscow. (As cited in Papirmeister et al., 1991)

- Sulzberger, M.B., R.L. Bauer, A. Kanof and C. Lowenberg. 1945. Skin sensitization to vesicant agents of chemical warfare. In: *Fasciculus on Chemical Warfare Medicine*, vol. 3, pp.16-66.
- Taher, A.A. 1992. Cleft lip and palate in Tehran. *Cleft Palate Craniofacial Journal* 29:15-16.
- Takehima, Y, K. Inai, W.P. Bennet, et al. 1994. P53 mutations in lung cancers from Japanese mustard gas workers. *Carcinogenesis* 15:2075-2079.
- Thomsen, A.B., J. Eriksen and K. Smidt-Nielsen. 1998. Chronic neuropathic symptoms after exposure to mustard gas: A long-term investigation. *J. Amer. Acad. Dermatol.* 39:187-190.
- Travis, C., S.A. Richter, E.A.C. Crouch, R. Wilson and E.D. Klema. 1987. Cancer risk management: a review of 132 federal regulatory decisions. *Environ. Sci. Technol.* 21:415-420.
- Uhde, G., Dunphy, E.B. 1944. The Effect of Oily Drops on Eyes Exposed to Mustard Vapor. No. Z.8307. Military Intelligence Division. Porton Down, UK.
- Urbanetti, J.S. 1988. Battlefield chemical inhalation injury. In: *Pathophysiology and Treatment of Inhalation Injuries*, J. Luke, ed. Marcel Dekker, NY. (As cited in Papirmeister et al., 1991)
- USEPA. 1984. *Pesticide Assessment Guidelines. Subdivision F. Hazard Evaluation: Human and Domestic Animals*, revised edition. PB86-108958. Office of Pesticide Programs, Washington, DC.
- USEPA. 1986a. Guidelines for Carcinogen Risk Assessment. *Federal Register* 51:33992-34003.
- USEPA. 1986b. *Superfund Public Health Manual*. EPA/540/1-86/066. Office of Emergency and Remedial Response, Washington, DC.
- USEPA. 1989. *Risk Assessment Guidance for Superfund: Volume 1 Human Health Evaluation Manual (Part A)*. EPA/540/1-89/002. Office of Emergency and Remedial Response, Washington, DC.
- USEPA. 1991. *Upper-bound Quantitative Cancer Risk Estimate for Populations Adjacent to Sulfur Mustard Incineration Facilities*. EPA/600/8-91/053. Office of Research and Development, U.S. Environmental Protection Agency, Washington, DC.
- USEPA. 1994. *Methods for the Derivation of Inhalation Reference Concentrations and Application of Inhalation Dosimetry*. EPA/600/8-90/066F. Office of Research and Development, Washington, DC.
- USEPA. 1996. Proposed Guidelines for Carcinogen Risk Assessment. Office of Research and Development, Washington, D.C. EPA/600/P-92/003C.
- USEPA. 1998. *Health Effects Test Guidelines, OPPTS 870.4100, Chronic Toxicity*. EPA 712-C-98-210. Prevention, Pesticides and Toxic Substances, Washington, DC.
- Venitt, S. 1968. Interstrand cross-links in the DNA of *Escherichia coli* B/r and B_{s-1} and their removal by the resistant strain. *Biochem. Biophys. Res. Commun.* 31:355-360.

- Vijayaraghavan, R. 1997. Modifications of breathing pattern induced by inhaled sulphur mustard in mice. *Arch. Toxicol.* 71:157-164.
- Wada, S., Y. Nishimoto, M. Miyashashi, et al. 1962a. Review of Okuno-jima poison gas factory regarding occupational environment. *Hiroshima J. Med. Sci.* 11:75-80.
- Wada, S., Y. Nishimoto, M. Miyashashi, S. Katsuta, M. Nishiki, A. Yamada, S. Tokuoka, H. Umisa, and M. Nagal, 1962b. Malignant respiratory tract neoplasms related to poison gas exposure. *Hiroshima J. Med. Sci.* 11:81-91.
- Wada, S., Y. Nishimoto, M. Miyashashi, et al. 1968. Mustard gas as a cause of respiratory neoplasm in man. *The Lancet*, June 1, 1968, pp. 1161-1163.
- Walker, I.G. and C.J. Thatcher. 1968. Lethal effects of sulfur mustard on dividing mammalian cells. *Radiat. Res* 34:110-127.
- Ward, D.M., N.M. Anson, P.A. Parent, and E.H. Enquist. 1966. *Sulfur Mustard and Analogous Compounds as Special Purpose Agents (U)*. 12-33 EASP 100-7R1. Aberdeen Proving Ground, MD. (As reported in Rosenblatt et al. 1975).
- Warthin, A.S., C.V. Weller and R.G. Herrman. 1918. The ocular lesions produced by dichlorethyl-sulphide (mustard gas). *J. Lab. Clin. Med.* 4:785-832.
- Waters, M.D., N.E. Garrett, C.M. CovonedeSerres, B.E. Howard, and H.F. Stack. 1983. Genetic toxicology of some known or suspected human carcinogens. In: *Chemical Mutagens, Principles and Methods for their Detection*, F.J. de Serres, ed., vol. 8. pp. 261-341. Plenum Press, New York.
- Watson, A.P., T.D. Jones, and G.D. Griffin. 1989. Sulfur mustard as a carcinogen: Application of relative potency analysis to the chemical warfare agents H, HD, and HT. *Reg. Toxicol. Pharmacol.* 10:1-25.
- Watson, A.P. and G.D. Griffin. 1992. Toxicity of vesicant agents scheduled for destruction by the chemical stockpile disposal program. *Environ. Health Perspect.* 98:259-280.
- Weiss, A. and B. Weiss. 1975. Karzinogenese durch Lost-Exposition beim Menschen, ein wichtiger Hinweis fur die alkylantien-Therapie. *Dtsch. med. Wschr.* 100:919-923 (as cited in IARC 1975)
- WHO (World Health Organization). 1970. Chemical Agents. In: *Health Aspects of Chemical and Biological Weapons*. World Health Organization, Geneva. (pp. 23-31)
- Wils, E.R.J., A.G. Hulst and J. Van Laar. 1988. Analysis of thiodiglycol in urine of victims of an alleged attack with mustard gas. Part. II. *J. Anal. Toxicol.* 12:15-19.
- Wulf, H.C., A. Aasted, E. Darre and E. Niebuhr. 1985. Sister chromatid exchanges in fishermen exposed to leaking mustard gas shells. *The Lancet*, March 25, 1985, pp. 690-691.
- Yamada, A., F. Hirose and M. Miyashashi. 1953. An autopsy of bronchial carcinoma found in a patient succumbed to occupational mustard gas poisoning. *Gann* 44:216-219. (as cited in IARC, 1975)

Yamada, A., F. Hirose, M. Nagai and T. Nakamura. 1957. Five cases of cancer of the larynx found in persons who suffered from occupational mustard gas poisoning. *Gann* 48:366-368. (as cited in IARC, 1975).

Yamada, A. 1963. On the late injuries following occupational inhalation of mustard gas, with special references to carcinoma of the respiratory tract. *Acta Pathologica Japonica* 13:131-155.

Yamada, A. 1974. Patho-anatomical studies on occupational poisoning. *Tr. Soc Path. Jap* 63:17-61. (as cited in Inada et al., 1978)

Yamakido, M., Y. Nishimoto, T. Shigenobu, K. et al. 1985. Study of the genetic effects of sulfur mustard gas on the former workers of Ohkunojima poison gas factory and their offspring. *Hiroshima J. Med. Sci.* 34:311-322.

Yamakido, M., J. Yanagida, S. Ishioka, S. Matsuzaka, S. Hozawa, M. Takaishi, T. Inamizu, M. Akiyama and Y. Nishimoto. 1986a. Immune functions of former poison gas workers. I. Mitogenic response of lymphocytes and serum factors. *Hiroshima J. Med. Sci.* 35:117-126.

Yamakido, M., J. Yanagida, S. Ishioka, S. Matsuzaka, S. Hozawa, M. Takaishi, T. Inamizu, M. Akiyama and Y. Nishimoto. 1986b. Immune functions of former poison gas workers. II. Lymphocyte subsets and interleukin 2 production. *Hiroshima J. Med. Sci.* 35:127-134.

Yamakido, M., S. Ishioka, K. Hiyama and A. Maedo. 1996. Former poison gas workers and cancer: Incidence and inhibition of tumor formation by treatment with biological response modifier N-CWS. *Environ. Health Perspect.* **104**:485-488.

Yanagida, J., S. Hozawa, S. Ishioka, et al. 1988. Somatic mutation in peripheral lymphocytes of former workers at the Okunojima poison gas factory. *Jap. J. Cancer Res.* **79**:1276-1283.

Young, L., J.A. McCarter, M. Edson and E. Estok. 1944. *Biochemical Experiments with Mustard Gas Prepared from Radioactive Sulfur. V. The Systemic Distribution of ³⁵S at Different Times after Application of Radioactive Mustard Gas to the Skin of the Rat.* University of Toronto, Report No. 17, C.P. 75. (As cited in Anslow and Houck, 1946).

Young, R.A., D.M. Opresko, A.P. Watson, et al. 1999. Deriving toxicity values for organophosphate nerve agents: A position paper in support of the procedures and rationale for deriving oral RfDs for chemical warfare nerve agents. *Human Ecol. Risk Assess.* 5:589-634.

GLOSSARY

Airborne Exposure Limits (AELs)	<p>Workplace: Atmospheric concentration levels (mg/m^3) for the workplace, which would not result in adverse health effects, based upon an 8 hr TWA for unprotected workers who may be repeatedly exposed for 8 hr/day, 40 hr/week, for a working lifetime.</p> <p>General Population: Atmospheric concentration levels (mg/m^3) allowable for the general population (including sensitive subpopulations) for indefinite, unprotected lifetime exposure where no adverse health effects are expected as a result of exposure. The existing general population AEL (DHHS, 1988) was expressed as a 72 hr TWA only to reflect sampling requirements at the time of the original CDC publication (DHHS, 1988).</p>
Acute Toxicity	Toxic effects resulting from a single exposure to a toxicant occurring within a 24 hr time frame from the exposure period.
Adverse Effect	Refers to either biochemical change, functional impairment, or pathologic lesion which impairs performance and reduces the ability of an organism to respond to additional challenge.
Ceiling Limit	The concentration that should not be exceeded during any part of the working exposure. In conventional industrial hygiene practice, if instantaneous monitoring is not feasible, then the Ceiling Limit can be assessed by sampling over a 15-minute period except for those substances that may cause immediate irritation when exposures are short.
Critical Effect	The first adverse effect or its known precursor that occurs as dose rate increases.
General Population Limit (GPL)	Airborne exposure level (AEL) for long-term general population exposure expressed as an atmospheric concentration.
Immediate versus Delayed Toxicity	Immediate effects occur or develop rapidly after a single administration of a substance, while delayed effects are those that occur after a lapse of some time.
IDLH	Immediately dangerous to life or health (IDLH) concentrations represent the

	maximum concentration from which, in the event of respirator failure, one could escape within 30 min without a respirator and without experiencing any escape impairing (e.g. severe eye irritation) or irreversible health effects.
Local versus Systemic Toxicity	Local effects refer to those that occur at the site of entry (e.g., respiratory tract, eyes) of a toxicant into the body; systemic effects are those that are elicited after absorption and distribution of the toxicant from its entry point to a distant site.
Lowest Observed Adverse Effect Level (LOAEL)	The lowest exposure level at which there are statistically or biologically significant increases in frequency or severity of adverse effects between exposed population and its appropriate control group.
No Observed Adverse Effect Level (NOAEL)	The exposure level at which there are no statistically or biologically significant increases in the frequency or severity of adverse effects between the exposed population and its appropriate control; some effects may occur at this level, but they are not considered as adverse, nor precursors to specific adverse effects. In an experimental study in which several NOAELs are determined, the regulatory focus is primarily on the NOAEL seen at the highest dose. This leads to the common usage of the term NOAEL to mean the highest exposure without adverse effect for a specific study.
Reference Concentration (RfC)	An estimate (with uncertainty spanning perhaps an order of magnitude) of a continuous inhalation exposure to the human population (including sensitive subgroups) that is likely to be without appreciable risk of deleterious non-cancer effects during a lifetime. The EPA has adapted the reference dose method for oral exposures to set airborne exposure levels for health effects other than cancer.
Severity of Effect	The degree to which an effect changes and impairs the functional capacity of an organ system.
Short-Term Exposure Limit (STEL)	The concentration to which workers can be exposed continuously for a short period of time without suffering from 1) irritation, 2) chronic or irreversible tissue damage, or 3) narcosis of sufficient degree to increase the likelihood of accidental injury, impair self-rescue or materially reduce work efficiency, and provided that the daily TLV-TWA s not exceeded. The STEL category of the TLV-TWA was developed by the American Conference of Governmental Industrial Hygienists (ACGIH) to define a 15 minute time weighted average (TWA) exposure which should not be exceed at any time during a workday even if the 8 hr TWA is within the threshold limit value (TLV) TWA.

	Exposures above the TLV-TWA up to the STEL should not be longer than 15 min and should not occur more than four times per day. There should be at least 60 min between successive exposures in this range.
Threshold	A dose level below which a response is unlikely, because homeostatic, compensatory and adaptive mechanisms in the cell or organism protect against toxic effects.
TWA	Time-weighted average concentration.
Uncertainty Factor (UF)	One of several factors used in operationally deriving the Reference Dose (RfD) or Reference Concentration (RfC) from experimental data. UFs are intended to account for 1) the variation in sensitivity among the members of the general human population; 2) the uncertainty of extrapolating animal data to humans; 3) the uncertainty in extrapolating from data obtained in a study that is of less than-lifetime exposure; 4) the uncertainty in using LOAEL data rather than NOAEL data; and 5) the inability of a single study to address adequately all possible adverse outcomes in man.
Worker Population Limit (WPL)	Airborne-exposure level (AEL) for long-term occupational worker population exposure expressed as an atmospheric concentration.

SOURCE: *Glossary of Terms for Chemical Agents and Chemical Defense Equipment*. TG 204. U.S. Army Center for Health Promotion and Preventive Medicine. Aberdeen Proving Ground, MD (Dec. 1994).

APPENDIX A

LOGISTIC AND PROBIT ANALYSIS FOR ESTIMATION OF SULFUR MUSTARD STEL

STEL
Log 10

The LOGISTIC Procedure

Data Set: WORK.STEL
Response Variable (Events): R
Response Variable (Trials): N
Number of Observations: 41
Link Function: Logit

Response Profile

Ordered Value	Binary Outcome	Count
1	EVENT	29
2	NO EVENT	70

Model Fitting Information and Testing Global Null Hypothesis BETA=0

Criterion	Intercept Only	Intercept and Covariates	Chi-Square for Covariates
AIC	121.741	82.990	
SC	124.336	90.775	
-2 LOG L Score	119.741	76.990	42.751 with 2 DF (p=0.0001) 31.596 with 2 DF (p=0.0001)

Analysis of Maximum Likelihood Estimates

Variable	DF	Parameter Estimate	Standard Error	Wald Chi-Square	Pr > Chi-Square	Standardized Estimate	Odds Ratio	Variable Label
INTERCPT	1	-0.6620	0.4023	2.7074	0.0999			Intercept
LCONC	1	-2.3050	0.5557	17.2029	0.0001	-1.246497	0.100	
EXPTIME	1	-0.0176	0.0105	2.8373	0.0921	-1.433984	0.983	EXPTIME

Association of Predicted Probabilities and Observed Responses

Concordant = 86.6%	Somers' D = 0.743
Discordant = 12.2%	Gamma = 0.753
Tied = 1.2%	Tau-a = 0.311
(2030 pairs)	c = 0.872

STEL
Log 10

Probit Procedure

Iter	Ridge	LogLikelihood	INTERCPT	Log10 (CONC)	EXPTIME
0	0	-68.62157087543	0	0	0
1	0	-43.30541185751	-0.269593912	-0.671841647	-0.003271695
2	0	-39.14144748596	-0.401094053	-1.078315429	-0.005408851
3	0	-38.32040614588	-0.459579999	-1.298895914	-0.00732561
4	0	-38.14207346356	-0.438285886	-1.349387007	-0.009253703
5	0	-38.11088010156	-0.412400176	-1.354159395	-0.010496029
6	0	-38.11003598349	-0.407592261	-1.355003028	-0.010743127
7	0	-38.11003522656	-0.407446453	-1.355030063	-0.010750808

STEL
Log 10

Probit Procedure

Data Set =WORK.STEL
Dependent Variable=R
Dependent Variable=N
Number of Observations= 41
Number of Events = 29 Number of Trials = 99

Log Likelihood for NORMAL -38.11003523

Last Evaluation of the Gradient

INTERCPT	Log10 (CONC)	EXPTIME
0.000000969	-0.000000561	0.000204

Last Evaluation of the Hessian

	INTERCPT	Log10 (CONC)	EXPTIME
INTERCPT	36.531193	-12.533047	871.567722
Log10 (CONC)	-12.533047	15.395119	-370.047720
EXPTIME	871.567722	-370.047720	46264

Goodness-of-Fit Tests

Statistic	Value	DF	Prob>Chi-Sq
Pearson Chi-Square	32.7956	37	0.6665
L.R. Chi-Square	40.3507	37	0.3244

Response Levels: 2 Number of Covariate Values: 40

NOTE: Since the chi-square is small ($p > 0.1000$), fiducial limits will be calculated using a t value of 1.96.

STEL
Log 10

Probit Procedure

Variable	DF	Estimate	Std Err	ChiSquare	Pr>Chi	Label/Value
INTERCPT	1	-0.4074465	0.238202	2.925833	0.0872	Intercept
Log10 (CON)	1	-1.3550301	0.30293	20.00843	0.0001	CONC
EXPTIME	1	-0.0107508	0.006323	2.891226	0.0891	EXPTIME

STEL
Log 10

Probit Procedure
Probit Analysis on Log10(CONC)

Probability	Log10(CONC)	95 Percent Fiducial Limits	
		Lower	Upper
0.01	1.04748	0.53019	2.25558
0.02	0.84630	0.37961	1.90819
0.03	0.71866	0.28244	1.68943
0.04	0.62264	0.20824	1.52596
0.05	0.54454	0.14704	1.39384
0.06	0.47806	0.09423	1.28210
0.07	0.41977	0.04730	1.18474
0.08	0.36758	0.00472	1.09814
0.09	0.32012	-0.03452	1.01990
0.10	0.27643	-0.07114	0.94837
0.15	0.09553	-0.22895	0.65842
0.20	-0.04824	-0.36383	0.43744
0.25	-0.17158	-0.48870	0.25701
0.30	-0.28235	-0.60979	0.10393
0.35	-0.38498	-0.73064	-0.02928
0.40	-0.48238	-0.85342	-0.14758
0.45	-0.57661	-0.97960	-0.25464
0.50	-0.66935	-1.11034	-0.35345
0.55	-0.76208	-1.24680	-0.44654
0.60	-0.85632	-1.39043	-0.53615
0.65	-0.95371	-1.54320	-0.62446
0.70	-1.05635	-1.70803	-0.71369
0.75	-1.16712	-1.88938	-0.80651
0.80	-1.29046	-2.09459	-0.90660
0.85	-1.43423	-2.33708	-1.01998
0.90	-1.61512	-2.64584	-1.15897
0.91	-1.65881	-2.72089	-1.19206
0.92	-1.70628	-2.80260	-1.22784
0.93	-1.75847	-2.89264	-1.26699
0.94	-1.81676	-2.99340	-1.31050
0.95	-1.88324	-3.10858	-1.35987
0.96	-1.96134	-3.24421	-1.41758
0.97	-2.05736	-3.41133	-1.48812
0.98	-2.18500	-3.63407	-1.58132
0.99	-2.38617	-3.98620	-1.72715

STEL
Log 10

Probit Procedure
Probit Analysis on CONC

Probability	CONC 95 Percent Fiducial Limits		
		Lower	Upper
0.01	11.15516	3.38995	180.12562
0.02	7.01940	2.39671	80.94590
0.03	5.23192	1.91620	48.91372
0.04	4.19414	1.61526	33.57062
0.05	3.50380	1.40294	24.76505
0.06	3.00650	1.24231	19.14681
0.07	2.62889	1.11508	15.30186
0.08	2.33121	1.01094	12.53549
0.09	2.08986	0.92358	10.46891
0.10	1.88984	0.84891	8.87914
0.15	1.24604	0.59028	4.55424
0.20	0.89487	0.43268	2.73804
0.25	0.67363	0.32456	1.80721
0.30	0.52198	0.24559	1.27036
0.35	0.41211	0.18594	0.93479
0.40	0.32932	0.14015	0.71190
0.45	0.26509	0.10481	0.55636
0.50	0.21412	0.07756	0.44315
0.55	0.17295	0.05665	0.35765
0.60	0.13921	0.04070	0.29097
0.65	0.11125	0.02863	0.23743
0.70	0.08783	0.01959	0.19334
0.75	0.06806	0.01290	0.15613
0.80	0.05123	0.00804	0.12399
0.85	0.03679	0.00460	0.09550
0.90	0.02426	0.00226	0.06935
0.91	0.02194	0.00190	0.06426
0.92	0.01967	0.00158	0.05918
0.93	0.01744	0.00128	0.05408
0.94	0.01525	0.00102	0.04892
0.95	0.01308	0.0007788	0.04366
0.96	0.01093	0.0005699	0.03823
0.97	0.00876	0.0003879	0.03250
0.98	0.00653	0.0002322	0.02622
0.99	0.00411	0.0001032	0.01874