8 Chronic Effects of Acute, Low-Level Exposure to the Chemical Warfare Agent Sulfur Mustard*

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I. INTRODUCTION

Chemical warfare agents have been around for at least 4000 years and probably were originally used as poisons on individuals. The use of chemical weapons dates from at least 423 B.C. when allies of Sparta in the Peloponnesian War took an Athenian-held fort by directing smoke from lighted coals, sulfur, and pitch through a hollowed-out beam into the fort. Other conflicts during the succeeding centuries saw the use of smoke and flame. During the seventh century A.D., the Greeks invented "Greek fire,"

*The opinions or assertions contained herein are the private views of the authors, and are not to be construed as reflecting the view of the Department of the Army or the Department of Defense.

a combination probably of rosin, sulfur, pitch, naphtha, lime, and saltpeter that floated on water and was particularly effective in naval operations. During the fifteenth and sixteenth centuries, Venice employed unspecified poisons in hollow explosive mortar shells and sent poison chests to its enemy to poison wells, crops, and animals.[1–3] Finally, World War I and the Iran-Iraq War saw the advent of modern chemical warfare.

Mustard has been stockpiled in the arsenals of various countries since it was first used on July 12, 1917, when the Germans fired shells containing mustard at British troops entrenched near Ypres, Belgium. When a single agent was identified as the source of injury, it was estimated that mustard caused about 80% of the chemical casualties in World War I; other agents such as chlorine and phosgene caused the remaining 20%. The British had 180,983 chemical casualties; the injuries of 160,970 (88%) were caused solely by mustard. Of these casualties, 4,167 (2.6%) died. Of the 36,765 single-agent United States (U.S.) chemical casualties, the injuries of 27,711 (75%) were caused solely by mustard. Of the casualties who reached a medical treatment facility, 599 (2.2%) died. Just as disconcerting was the fact that mustard survivors required lengthy hospitalizations: the average length of stay was 42 days.[4]

Since the first use of mustard as a military weapon, there have been a number of isolated incidents in which it was reportedly used. In 1935, Italy probably used mustard against Abyssinia (now Ethiopia); Japan allegedly used mustard against the Chinese from 1937 to 1944; and Egypt was accused of using the agent against Yemen in the mid-1960s.

Chemical agents were not used during World War II. It is thought that Germany did not use mustard because Hitler had been a mustard victim during World War I and was loathe to use it.[4]

Sidell and Hurst have described the long-term effects produced from acute symptomatic clinical dose exposure to mustard, but less is known about the clinical effects from chronic, sometimes symptomatic, low-dose exposure.[5,6] Acute is defined here as an exposure lasting less than 24 h. The term chronic refers to an exposure lasting for days, weeks, months, and even years. Clinical means producing a recognizable illness directly related to mustard exposure. Symptomatic means having either the acute or chronic clinical illness produced by sulfur mustard. Asymptomatic, of course, is without symptoms at all.

The argument can be made that acute, subclinical asymptomatic injury causing long-term effects does not exist. The pros and cons will be considered in this chapter, and parallels will be drawn. Certainly chronic, subclinical asymptomatic exposures do exist and there are parallels to other harmful situations. Obviously, workers in the manufacture of sulfur mustard, who were asymptomatic for part or all of their employment, fall into this category.

II. CLINICAL EFFECTS OF SULFUR MUSTARD

The organs most commonly affected by mustard are the skin, eyes, and airways: the organs which mustard contacts directly. After a substantial amount of mustard has been absorbed through the skin or inhaled, the hemopoietic system, gastrointestinal

tract, and CNS are also damaged. Mustard may also affect other organs but rarely do these produce clinical effects.[4] After an asymptomatic latent period of hours, mustard causes erythema and blisters on the skin. This response ranges in severity from mild redness resembling sunburn to severe third-degree burns. Eye damage ranges from mild irritation-conjunctivitis, to corneal opacity, to perforation of the eye, and blindness. In the lung, the injury extends from mild upper respiratory signs to marked airway damage, bronchitis, and pneumonia. On rare occasions, acute laryngospasm can result in rapid death. Gastrointestinal effects vary from nausea and vomiting to severe hemorrhagic diarrhea. In the bone marrow, severe stem cell suppression can result in profound pancytopenia. In the CNS, at least in laboratory animals, seizures and death have been produced at high concentration exposures. The worst possible outcome from mustard exposure is death. However, mortality from mustard is uncommon. Less than 5% mortality from mustard gas was observed in allied troops in World War I.

Laboratory animal studies have shown that mustard is mutagenic and carcinogenic, and thus, it is not surprising that it is carcinogenic in man.[7–9] Both Morgenstern et al. and Buscher and Green emphasize that chronic low-dose exposure over months to years in occupationally exposed workers leads to chronic bronchitis, bronchial asthma, hoarseness, aphonia, and hypersensitivity to smoke, dust, and fumes.[10,11] Such individuals typically show persistent disability, with increased susceptibility to respiratory tract infections and evidence of bronchitis and bronchiectasis.[10–12]

All human studies dealing with chronic mustard disease processes are retrospective and fraught with the problems inherent in retrospective studies. These problems include bias in the sampling populations; lack of epidemiological controls from the effects of smoking, lifestyle, race, gender, age, or exposure to other chemicals; differential quality of available health care; and incorrect diagnosis.[12] These limitations make absolute interpretation of the studies difficult.

A. Carcinogenesis

Mustard is an alkylating agent similar to drugs that have been used in cancer chemotherapy, such as nitrogen mustards, Cytoxan, and cis-platin. Since DNA is one of mustard's most sensitive targets, it is not surprising that carcinogenesis and radiomimetic effects are seen.[5]

Human data on the carcinogenicity of mustard are from (a) battlefield exposures, (b) accidents, and (c) workers in chemical factories. Both British and American studies have investigated the increased incidence of pulmonary carcinoma arising from World War I battlefield exposure. All are difficult to interpret, owing to the lack of controls for age, chronic pulmonary disease, cigarette smoking, and other factors that might affect the outcome.[13–15]

In contrast to battlefield exposures, studies of factory workers involved in the production of mustard have shown a definite link between prolonged exposure to low doses of mustard and cancer.[12] Several studies have provided evidence of an increased risk of respiratory tract cancers in factory workers.[6,16–20] Easton et al. found a 45% increase in death due to lung cancer, a 170% increase in death from cancer of the larynx, and a 450% increase in death from cancer of the pharynx, compared with

expected deaths in the general population.[17] The risks from cancer of the pharynx and lung were significantly related to the duration of employment at the factory.

B. Chronic Pulmonary Disease

Inhalation of mustard vapor primarily affects the laryngeal and tracheobronchial mucosa.[12] Evidence exists to suggest that mustard inhalation causes sustained respiratory difficulties even after the acute lesions have healed. Clinical follow-ups on 200 Iranian soldiers who were severely injured by mustard during the Iran-Iraq War indicate that about one third had experienced persistent respiratory effects 2 years or more after initial exposure. Reported problems included chronic bronchitis, asthma, rhinopharyngitis, tracheobronchitis, laryngitis, recurrent pneumonia, bronchiectasis, and, in some cases, severe, unrelenting tracheobronchial stenosis.[21–25]

Of the British soldiers exposed to mustard in World War I, 12% were awarded disability compensation for respiratory disorders that were believed to be due to mustard exposures during combat.[26]

Little contemporary information regarding the pathogenesis of the respiratory lesions is available, and few data from people or animals exposed to nonlethal concentrations of mustard vapor exist. Even fewer studies investigate the histopathology of the recovery process in animals exposed to mustard.[9] However, two studies conducted during World War I suggest that low-level exposure or survivable exposures in dogs and rabbits may produce scar tissue following small ulcerations in the trachea and larynx, causing contractions of these areas.[27,28] The more severe respiratory tract lesions described in animals exposed to mustard vapor appear to be quite similar in type and location to those described in humans.[12]

C. Chronic Eye Disease

Individuals who sustain acute ocular injury due to high-dose mustard exposure may experience difficulties even after the initial effects of the injury have subsided.[29–32] Recurrent or persistent corneal ulceration can occur after latent periods of 10 to 25 years. Chronic conjunctivitis and corneal clouding may accompany this delayed keratopathy.[31–32] Anecdotal accounts suggest that low-dose exposure also causes increased sensitivity to later exposure to mustard, although the existence of increased sensitivity is difficult to substantiate with available scientific evidence.[12, 33]

D. Scarring, Pigmentation Changes, and Cancer of Epithelial Surfaces

Skin cancer occurring at the site of old scar formation is an acknowledged biological phenomenon.[34,35] Cutaneous cancers resulting from acute mustard exposure usually localize in scars, whereas those caused by chronic exposure can occur on any exposed site.[36]

In a prospective study of delayed toxic effects from mustard exposure, Balali-Mood followed a group of Iranian solders exposed to mustard gas during the Iran-Iraq War.[24] After 2 years, 41% of the exposed victims were experiencing pigmentary disorders.

In the absence of melanocyte destruction, hyperpigmentation predominates. If melanocytes are locally destroyed, and inward migration from destroyed adnexal structures does not occur, depigmentation predominates.[5]

In its study of mustard and Lewisite effects, the Institute of Medicine concluded that, following mustard exposure:

- The evidence indicates a causal relation between acute, severe exposure to mustard agents and increased pigmentation and depigmentation in human skin.
- Acute and severe exposure can lead to chronic skin ulceration, scar formation, and the development of cutaneous cancer.
- Chronic exposure to minimally toxic and even subtoxic doses can lead to skin pigmentation abnormalities and cutaneous cancer.[9]

E. Central Nervous System

Excitation of the CNS after mustard exposure, resulting in convulsions and followed by CNS depression, has been reported by the U.S. Army.[37] Convulsions and cardiac irregularities appear to occur only after extremely acute, high doses, which are probably attainable only in laboratory settings.[12, 38] Mustard casualties of the Iran-Iraq War did not display severe CNS or cardiac abnormalities.[21]

F. Summary for Symptomatic Exposures

The organs most commonly affected by mustard are the skin, eyes, and airways; the organs mustard comes in direct contact with. After a substantial amount of mustard has been absorbed through the skin or inhaled, the hemopoietic system, gastrointestinal tract, and CNS are also damaged. Mustard may also affect other organs, but rarely do these produce clinical effects.[4] After an asymptomatic latent period of hours, mustard causes erythema and blisters on the skin. This ranges in severity from mild redness resembling sunburn, to severe third-degree burns. Eye damage ranges from mild irritation-conjunctivitis to corneal opacity, or even perforation of the eye and blindness. In the lung, the injury ranges from mild upper respiratory signs to marked airway damages, bronchitis, and pneumonia. On rare occasion, acute laryngospasm can result in rapid death. Gastrointestinal effects range from nausea and vomiting to severe hemorrhagic diarrhea. And in the bone marrow, severe stem cell suppression can result in profound pancytopenia. In the CNS, at least in laboratory animals, seizures and death have been produced at high concentration exposures. The worst outcome from all these organs systems, except for possibly the eye, is death. Death, however, is not the usual outcome from mustard exposure.

Studies of English and Japanese mustard factory workers establish repeated symptomatic exposures to mustard over a period of years as a causal factor in an increased incidence of airway cancer. The association between a single exposure to mustard and airway cancer is not as well established as the association between one-time mustard exposure and other chronic airway problems, such as chronic

bronchitis (based on World War I data). In some cases, the long-term damage was probably a continuation of the original insult resulting from insufficient therapy in the pre-antibiotic era. Morgenstern et al. give the following graphic description of symptoms and injuries incurred by some of the mustard factory workers.

> Less widely known is the fact that many persons employed in the handling of mustard gas and exposed to small quantities of the vapor over a prolonged period of time may sustain damage to the respiratory mucosa, which may leave them partially or totally disabled. This statement is based on two and one-half years of observation in the medical department of an industrial plant where over 200 patients have been treated for both the acute symptoms and the residual effects of mustard gas exposure. The evolution of chronic mustard bronchitis may be traced as follows:
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> A young, white male previously engaged in farming or some other nonindustrial occupation with no history of any previous chronic lung disease goes to work on the mustard filing line. There is a varying concentration of mustard vapor in the air during a good part of the working day. After a period of time ranging anywhere from 3 weeks to 6 or 12 months he begins to show signs of definite irritation of the conjunctival and respiratory mucous membranes. He develops symptoms. He is given sick time off with his condition improving and returns to work. After a number of such episodes it becomes apparent that this man is not suitable for work in mustard and he is transferred out to another department free of toxic fumes.
>
> After removal from mustard, his eyes and throat gradually heal. The conjunctivitis recedes and the vision returns to normal. The sore throat and hoarseness subside. The sense of taste returns, but the sense of smell may remain impaired. The appetite improves and he regains some of his lost weight with overall improvement. But he remains troubled by a persistent hacking cough, which come in paroxysms. It is most common in the morning but also occurs on lying down at night. It is often precipitated by physical exertion or when the man walks from the cold into a warm room or comes into contact with fumes of smoke. The cough is productive of anywhere from a teaspoon to a cupful of white or yellow mucoid or mucopurulent sputum, which may have a foul odor on occasion. There may be a troublesome wheezing and chest tightness most marked during damp weather. The patient seems to be more susceptible to respiratory infections than he was prior to exposure to mustard and the infections tend to last longer. Definite clinical bronchiectasis may develop as a result of repeated attacks of acute infectious bronchitis. He is hypersensitive to fumes and dust of any kind. He may develop dyspnea on slight or moderate exertion and therefore cannot perform any arduous labor.[10]

This description of chronic bronchitis developing in factory workers in the setting of World War II is both accurate and quite convincing.

Several eye diseases, such as chronic conjunctivitis, appear after an acute, usually severe, insult to the eye. In particular, delayed keratitis has appeared more than 25 years after the acute, severe lesion. Similarly, skin scarring, pigment changes, and even cancer have either followed the initial wound as a continuation of the process (scarring) or later appeared at the site of the lesion.

The production of nonairway cancer by mustard has been demonstrated in animals, but scant evidence exists to implicate mustard as a causative factor in nonairway cancer in humans.[5]

III. ACUTE SUBCLINICAL EXPOSURE

We are not convinced that acute, asymptomatic injuries that result in clinical disease truly exist. It is conceivable that certain synergistic situations can develop, such as would happen if co-factors or preexisting conditions (immunosuppression, genetic deficiency, or an additional chronic subclinical exposure) were triggered by some otherwise uneventful insult. This is certainly an unknown for mustard exposure at this time. The best that can be done is to draw analogies to other circumstances.

A. CARCINOGENESIS

Genotoxic substances usually have a direct effect on DNA and are occasionally effective after a single exposure. This helps to explain why they are frequently carcinogenic at subtoxic doses. These toxic compounds often act in a cumulative manner and synergistically with other DNA-reactive carcinogens. They usually produce neoplasms in more than one target organ and have a variable latency.[39]

Strong promoters also possess weak intrinsic carcinogenicity. This experimental evidence comes from two test systems: (1) continued high-level administration of promoters such as croton oil to mouse skin or (2) oral administration of DDT or phenobarbarbital to rats. Both systems yield a small but definite crop of benign and malignant neoplasms in the absence of any obvious genotoxic carcinogen. An explanation is needed because promoters, by definition, do not have intrinsic properties of altering the genetic apparatus. One example has supplied an explanation that could apply to the others: when croton oil was applied to the skin of mice, it appeared to induce by itself a high incidence of papillomas and carcinomas.[40] Careful analysis revealed that the mice used were purchased from a supplier who housed the mice in creosoted cages. Thus, the mice had been exposed to genotoxic carcinogens in the creosote prior to the application of croton oil. Similarly, the weak carcinogenicity of DDT or phenobarbital may stem from prior exposure of the animals to small amounts of carcinogens, possibly mycotoxins or certain nitrosamines in the diet.[41–43]

In mice, chemical enzyme-inducing substances have produced liver tumors, but were not found to be genotoxic using *in vitro* testing. Such chemicals have shown promoting activity in these systems.[44–46] The liver of these animals appears to respond as if a DNA gene structure change has occurred. The role mycotoxins or nitrosamines play in the diet of these animals is open for speculation. Complex polychlorinated aliphatic and cyclic hydrocarbons also fall into the class of enhancing substances.[47,48] They exhibit a nonlinearity in their dose-time response curves that differs from the genotoxic carcinogens.[49]

The mechanism of promotion is subject to scientific research and considerable conjecture. The probability of multiple mechanisms is strong. A sequence of steps may be involved, leading to proliferation and differentiation. A parallel example might be the prostaglandin and cyclic nucleotide membrane effector systems. An early biochemical indicator of promotion is the induction of ornithine decarboxylase (ODC), and its presence has been used to discover new promoters.[50–55] Increased levels of ODC appear to be associated with increased liver cell proliferation and might be related to the mechanisms of the promotion.[56, 57]

Promoters and carcinogens involved in human cancer induction were first discovered through carefully conducted epidemiological studies, and then tested in animal models or in *in vitro* systems. Newer techniques use exfoliated cells or cells in culture to ascertain exposure to these genotoxic materials.[57–60]

Also, monoclonal antibody techniques may be able to trace carcinogen-macromolecular adducts in human tissues, verifying exposure to specific carcinogens.[61,62] The gold standard will be to identify human hazards and carcinogens before exposure, "an ounce of prevention is worth a ton of cure."[63–70]

B. Radiation

Ultraviolet light is a form of nonionizing radiation exposure to man, while X-rays are an ionizing radiation. Both forms have the effect of damaging DNA similar to sulfur mustard, especially in rapidly dividing cells. It may never be possible to determine whether an individual can go through life without manifesting symptoms of acute sun damage: erythema, blisters, pigmentation, etc. or, at the very least, mild erythema. But, the chronicity of exposure producing the skin damage and cancers is undeniable. It is the acute symptomatic vs. chronic asymptomatic mix of exposures that will always remain a mystery.

Sulfur mustard has been called "radiomimetic" primarily because it appears to target DNA in rapidly dividing cells. There is a latent onset to its acute clinical symptoms very much paralleling those of ultraviolet sunlight damage, and a delayed onset to pulmonary cancers from chronic exposure. It is well established that the consequence of lifelong exposure to sunlight significantly enhances the development of skin cancers in fairer skinned individuals. The chronic damage to fair-skinned individuals is seen as premature aging, pigmentary changes, solar elastosis, solar keratosis, basal and squamous cell cancers, and malignant melanoma.[71,72] The threshold for developing these lesions in man is multifactoral: heredity (skin type, individual's ability to repair DNA), environment, and lifestyle. Thus, the cumulative dose responsible for these affects has great variability. Dark-skinned individuals may never experience any of the clinical entities mentioned above while individuals with the genetic disorder, Xeroderma pigmentosa, will experience marked acceleration of sun-related skin damage and cancers. This is caused by the genetic defect, which impairs the DNA repair produced by ultraviolet damage.[73]

Likewise, it is well recognized that certain dyes, pigments, and drugs (i.e., tetracycline, psoralens, etc.) enhance sunlight and artificial light effects on skin. Existing genetic defects and specific drugs have not been identified that enhance the damage caused by sulfur mustard, but this possible synergism may be acute, subacute, or chronic.

Radiation-induced cancer is due to a nonlethal mutation of somatic cells. The latent period between irradiation and the development of cancer varies from 4 to 40 years, the average being 7 to 12 years. Even relatively low doses of X-rays increased the risk of cancer. Of school children epilated with 300 to 400 r of unfiltered 100-kV radiation, 1.6 percent had skin, thyroid, or parotid tumors 20 years later, while untreated control group showed only 0.2 percent such tumors. Apparently the mutated cells can survive 10 to 20 years before proliferating.[74]

Evidence that various derivatives of tar and oil cause squamous cell carcinoma of the skin is both environmental and experimental. Experimental production of skin cancer in rodents with the various carcinogenic hydrocarbons has been well demonstrated. In fact, the effect of a chemical on animal skin is currently regarded as the best method of testing the carcinogenicity of the chemical.[75]

IV. *IN VITRO* STUDIES OF SULFUR MUSTARD TOXICITY

Sulfur mustard is an alkylating agent that acts through cyclization of an ethylene group to form a highly reactive sulfonium electrophilic center. This reactive electrophile is capable of combining with any of the numerous nucleophilic sites present in macromolecules of cells. The products of these reactions are stable adducts which can modify the normal function of the target macromolecule. Since nucleophilic areas exist in peptides, proteins, RNA, DNA, and membrane components, extensive efforts have been underway to identify the most critical biomolecular reactions leading to mustard injury.

While the chemistry of mustard interactions with cellular components is well defined, the correlation of these interactions with injury has not been made. Over the past few decades, scientists have made major advances in understanding the cellular and biochemical consequences of exposure to mustard. While not the only target for alkylation by mustard, DNA is presumed to be an early reactant in the pathogenic cascades leading to the mustard lesion. Alkylation of nucleotides can result in apurinic site formation, disruption of normal DNA replication, activation of DNA repair pathways and eventually, to cytotoxic or mutagenic events. At high-exposure doses, such as those that lead to vesication *in vivo* or above 50 μM *in vitro,* the exposed cells sustain so much damage that they will die. The cells show activation of the nuclear enzyme poly(ADP-ribose) polymerase (PARP), disruption of cellular metabolism, loss of cell energetics, and total cellular breakdown. Many of these cells initially respond to the agent insult by activation of apoptotic death pathways, but in the absence of sufficient energy stores, quickly shift to a necrotic pattern of death. One could visualize this response as a cell suicide to eliminate the threat of long-term genotoxic sequelae such as mutation or cancer.

At low-dose exposure, the pattern could switch to one in which there is little to no immediate demonstration of injury but does set the stage for long-term consequences. As discussed previously, as a genotoxic agent, mustard can function in carcinogenesis in concert with, or playing the role of, promoter or inducer. The presence of nucleotide adducts or incorrect base replacement following DNA repair attempts coincidental with exposure to a second chemical insult could result in genesis of a cancer. But the question we wish to address is, "Can a low-dose, acute, asymptomatic exposure to mustard lead, by itself, to disease in later years?" Mustard is a mutagen and the mutation rate following *in vitro* exposure to cell culture systems has been studied. In the early 1970s, our laboratories reported that *in vitro* exposure of mouse lymphoma cells to submicromolar concentrations of mustard could increase the

reversion rate for asparagines independence. *In vivo,* using a host-mediated assay in mice, comparable reversion rates were seen with a single subcutaneous dose of 100 mg/kg.[76] In a concurrent study of chronic exposure over 37 weeks, mice were exposed in an inhalation chamber to 0.1 mg/m^3 for 6 h per day, 5 days/week. No statistically significant increase in reversion rate was detected.[77] It is also interesting to note that the bifunctional alkylating agent sulfur mustard is less mutagenic than many monofunctional agents presumably because the cross-links formed by sulfur mustard lead to death of the affected cell.

Besides genotoxicity, sulfur mustard is known to affect other cellular parameters. Concentrations of mustard above 50 μM result in a marked reduction of cellular NAD+, a critical glycolytic cofactor, within 4 h of *in vitro* exposure to human keratinocytes.[78] However, no alterations in NAD+, ATP, or mitochondrial dehydrogenase activity are seen at 10-fold less concentrations in this model. One of the central cellular enzymes involved in NAD+ turnover is the nuclear enzyme PARP. As mentioned earlier, this enzyme undergoes a large and rapid activation in human epithelial cells exposed *in vitro* to vesicating equivalent doses (i.e., >50 μM). At concentrations of mustard below 10 μM, however, a completely different pattern of PARP response is seen.[79] It appears that even though significant DNA damage is detected at these low doses, the repair response is not as aggressive and the net metabolic disruption is transient.[80]

As one studies the response to *in vitro* exposure to mustard in cell systems, there appears to be a threshold level above which death processes are initiated that are rapid and totally destructive to the cells. This appears to be in the range of 50–100 μM for most mammalian cells. If concentrations below 10 μM are studied, one can observe toxic processes occurring, but depending on the cell system employed, these are often reversible.

V. DOSE DEPENDENCY OF THE MUSTARD LESION

In 1946, Renshaw reviewed the understanding of the mechanisms of mustard injury to that point.[81] He defined three dose ranges based on μg of mustard fixed per cm^2 of human skin. From 0.1–1.0 μg fixed/cm^2 the result was "mild erythema, occasional vesication with a histology that showed hyperemia and edema *without sufficient epidermal injury to cause death of more than occasional isolated basal cells*" (italics added). For 1.0–2.5 μg fixed/cm^2, the result was moderate injury with routine blister formation, and at doses >2.5 μg fixed/cm^2, the resulting injury was described as severe with central necrosis and circumferential vesication. Furthermore, he went on to state that minimal reversible injury was seen at 0.1 μg fixed/cm^2. One can say, therefore, at exposures resulting in less than 0.1 μg mustard fixed per cm^2 of skin, the outcome will be minimal clinical symptoms and fully reversible changes with no long-term effects. This exposure level is less than one-tenth that required for full demonstration of vesication and very close to what we refer to as a mild erythematic exposure.

Finally, for many years, sulfur mustard was used topically in the treatment of psoriasis in the form known as Russian Ointment (0.005% mustard-vaseline). This

was also known as Psoriasin. In the 1970s, Illig reviewed the clinical information from these studies and evaluated the potential skin carcinogenicity and off-gassing problems associated with cutaneous exposure to low-dose mustard.[82,83] The following is a quote from his 1976 paper: "It is extremely improbable that the carcinogenic risk of the external S-mustard treatment is higher than that of a parenteral Methotrexate therapy, carried out at many clinics and in many cases over a period of many years, especially in the U.S.A.; it is rather to be expected that the carcinogenic risk in the case of external application of Psoriasin is also substantially lower over a longer period of time than in the Methotrexate treatment."

VI. SUMMARY

Genotoxic agents have the potential of long-term consequences, especially when synergistically coupled with promoters, immunosuppression or genetic deficiencies. Our contention, that acute subclinical asymptomatic injury causing long-term effects does not exist, is based on the following:

1. Lack of reliable clinical cases
2. *In vitro* observations
3. Renshaw's suggestion that, once the dose of applied mustard drops below that which yields observable, sustainable injury, no untoward consequences will become evident in the patient
4. More than 30 years' experience with Russian Ointment

Based on our scientific and medical experience, we should never-say-never, but the probability of chronic illness developing from an acute asymptomatic exposure to sulfur mustard appears to be extremely low.

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