

Therapeutics & Toxins News

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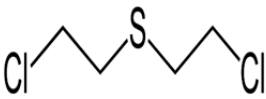
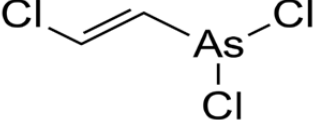
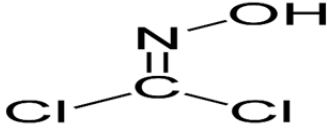
Part 1 – The Vesicants

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Nearly one century ago the first mass use of a chemical weapon occurred in the quaint Belgian city of Ypres resulting in an unprecedented German tactical success. As the unknown agent crept downwind and into the Allied trenches mass hysteria developed followed by a chaotic retreat that opened a several mile wide gap in the stalemated line. This 1915 event triggered the rapid escalation of chemical weapons usage, a race to develop more toxic compounds, and ushered in a new age of terror. Tragically, the master mind behind the initial release of this toxic chlorine gas cloud was the renowned scientist Fritz Haber, who was awarded the 1918 Nobel Prize in Chemistry for synthesizing ammonia. This article is the first in a two part series that focuses on the current chemical warfare agents. Initially we will discuss the vesicants and in a later issue complete our discussion with the nerve agents.

Vesicants are defined as those compounds that produce blistering of skin and mucous membranes (i.e. vesicles). Exposure normally occurs via inhalation or dermal absorption followed by injuries to the respiratory system, eyes and skin. The U.S. military categorizes three compounds as vesicants. They are, in order of importance, sulfur mustard (SM), lewisite and phosgene oxime. Often referred to as mustard gas, SM is actually a liquid at room temperature and a solid below 57.8 °F. Table 1 summarizes many of their key properties.

Table 1. Key Properties of the Vesicants

	Sulfur Mustard (SM)	Lewisite	Phosgene Oxime
Structure			
Melting Point (°C)	14.4	-18	34-40
LD50 (liquid), mg/kg	~100	40-50	Not Determined
Vapor LCt50* mg-min/m ³	1500	1200-1500	3200
Volatility (at 20°C) mg/m ³	610	4480	1800

* LCt50 is the concentration and time of exposure that is lethal to 50% of the population

Shortly after World War I chemical weapons were internationally condemned and their use banned by the Geneva Protocol (1). Despite this, all of the major powers in World War II had extensive stockpiles of ammunition laden with vesicants that were fortunately not utilized in battle. While the Germans had the most advanced chemical weapons technology at the time their reluctance to use them may have stemmed from Adolf Hitler's history of being a SM casualty. These agents received little notoriety for several decades apart from occasional use in limited regional conflicts. This radically changed in the 1980s when the Iraqi military extensively used SM during their war with Iran causing an estimated 100,000 casualties (2, 3). Today, the vesicants are again becoming a prominent military concern after unconfirmed reports of their theft from Iraqi stockpiles and for their potential to be used in a terrorist attack (4). Knowledge of these agents and the methods used to identify them is critical for the timely triage and treatment of vesicant casualties.

Sulfur Mustard, bis(2-chloroethyl) sulfide, C₄H₈Cl₂S, is the most widely used and successful chemical weapon ever utilized. Even though introduced late in World War I by the Germans in July 1917, it caused ~1 million casualties. This accounted for a staggering 80% of all chemical casualties (3). SM vapor is 5.4 times heavier than air and thus settles at ground level which made it an ideal weapon to use in trench warfare. It is a thioether with low volatility that is slow to dissolve in water and appears as a pale

yellow to dark brown oily liquid noted for smelling like garlic, mustard or horseradish (Figure 1).

Figure 1. World War II identification posters of SM and lewisite.



SM is a potent alkylating agent that initially acts through cyclization of an ethylene group to form a sulfonium ion. This highly reactive electrophile quickly reacts with any of the abundant nucleophilic sites present in the common macromolecules (i.e. proteins, nucleic acids, and lipids) forming stable adducts. These are detrimental to their normal function and eventually lead to the disruption of the epidermal-dermal junction. When visualized by light microscopy there is liquefaction necrosis of epidermal basal cell keratinocytes (5). It's exact mechanism of action is unknown but current hypotheses include irreversible DNA alkylation causing guanine cross-links that result in strand breaks, reaction with glutathione causing a loss of cellular protection against oxidative stress, dysregulation of calcium homeostasis, indirect inhibition of glycolysis and the activation of the hexose monophosphate shunt (2, 5). It seems likely that the exact mechanism will involve a combination of these and potentially yet undiscerned mechanisms (6). Regardless, SM is so highly reactive *in vivo* that within a few minutes it is no longer present.

SM is insidious in action, often causing casualties without any warning. After a latent period of 4-12 hours, erythema develops that often progresses to external and internal

blistering. A 10 mcg droplet will cause vesication (2). Warm, moist areas with thin skin like the axillae, neck, antecubital fossae, perineum and external genitalia are the most sensitive areas of the body. Additionally, SM causes acute conjunctivitis often followed by temporary blindness that resolves in a few weeks (Figure 2). It is primarily an incapacitating agent; exposures were lethal in only ~3% of the World War I casualties. The rare deaths generally occurred four or more days post-exposure usually due to pulmonary insufficiency from airway damage complicated by infection and sepsis. The LD50 for SM liquid on skin is ~100mg/kg or ~7.0 g for a person weighing 70 kg (2). This is around 1.5 metric teaspoons of liquid; an amount that could cover ~25% of an adult's body surface area. SM slowly degrades in a non-aqueous environment and can remain potent on clothing or hair for several hours, in soil for months, concrete for decades, and for nearly 100 years in sealed munitions. Declared stockpiles currently exist in the U.S., Russia, previous Soviet-bloc states and most likely North Korea. Syria recently disclosed a large stockpile and there has been a major international effort to quickly destroy this material.

Figure 2. World War I combat casualties blinded by SM



Interestingly, another form of mustard exists that was never used on the battlefield, **nitrogen mustard**. It had previously been observed that SM casualties often developed leukopenia. In 1942 classified research showed nitrogen mustards could be used to treat leukemia and lymphoma. After the war, these clinical findings were disclosed and Merck marketed a nitrogen mustard product for these indications. This marked the

beginning of cancer chemotherapy and for a while was the main treatment used until replaced by less cytotoxic compounds.

Lewisite, b-chlorovinyl dichloroarsine, $C_2H_2AsCl_3$, is an arsenic-based vesicant developed by the U.S. during World War I. Weapons grade lewisite appears as a yellow to brown, oily liquid that has a distinct odor of geranium blossoms. It is insoluble in water but soluble in most organic solvents (7). The first shipment was partially across the Atlantic Ocean when the armistice was signed, so the vessel and its contents were scuttled at sea. Confirmed use of lewisite in combat has never been verified, although there is some evidence to suggest the Japanese used it against the Chinese in the 1940s. It has similar physiologic effects to SM but is more potent and volatile. The initial symptoms of exposure occur immediately; there is stinging pain upon skin or eye contact. Inhalation results in coughing, pain and tightness in the chest, often followed by nausea and emesis (8). This is an important clinical distinction; lewisite is immediately painful while initial contact with SM is not. Erythema develops from 15-30 minutes post-exposure followed by blisters several hours later. A 14 mcg droplet of liquid will cause vesication (2). It was extensively stockpiled by the Soviet Union and has often been mixed with SM to lower the latter's freezing point and produce a more potent chemical weapon.

The exact mechanism of lewisite's action is unknown but likely involves glutathione depletion as well as arsenical binding and inhibition of critical enzymes containing thiol groups. Studies show one key factor is the inhibition of carbohydrate metabolism, probably through inactivation of the pyruvate dehydrogenase complex (9). Lewisite, unlike SM, doesn't produce immunosuppression but is more rapidly absorbed through human skin. It increases the permeability of blood vessels leading to a phenomenon called lewisite shock. Exposure to large amounts can cause protein and plasma leakage from the capillaries and subsequent hypotension and hemoconcentration (2).

Phosgene Oxime, $CHCl_2NO$, is a yellowish-brown liquid that is different than simple phosgene, which chiefly affects the respiratory system. It is not a true vesicant since it doesn't produce vesicles but instead forms solid lesions (2). Skin contact almost instantaneously produces erythema and urticaria that eventually results in extensive tissue necrosis. Its effect on the lungs and eyes are similar to the other vesicants and it has an intense, irritating odor. It is capable of penetrating through clothing and rubber more quickly than the other vesicants. While there are no reports of it being used in battle, it was stockpiled by the former Soviet Union and often mixed with other chemical warfare agents. It is the least studied of the vesicants and its mechanism of action is unknown.

Lab Analysis and Detection: There are no routine laboratory tests for any of the vesicants. Limited studies on SM casualties showed the presence of appreciable amounts of its harmless major urinary metabolite thiodiglycol and 1,1'-sulfonylbismethylthioethane (2, 7). The latter is a more specific urinary marker of SM since it is absent from normal human urine. Both GC-MS and LC-MS have been utilized to measure intact SM in urine and its major metabolites. In an aqueous environment lewisite undergoes rapid hydrolysis to the toxic metabolite 2-chlorovinylarsonous acid. This metabolite has been assayed in blood and urine by GC-MS, urinary arsenic excretion can also be monitored (2, 7).

A wide variety of detection equipment is currently available commercially and through the U.S. Department of Defense. During the Cold War many tests, detectors, and monitors were developed and successfully used. New technologies are constantly being developed that show increased sensitivity and specificity.

Treatments: In order to effectively prevent or decrease tissue damage exposed skin should be promptly decontaminated in 1-2 minutes with copious water, dilute hypochlorite solution or if available, a military kit like the M291 which contains individual decontamination pads. Current treatment strategies are designed to relieve symptoms, prevent or limit infection, and promote healing (2). Animal studies suggest N-acetyl cysteine (NAC) has therapeutic promise to treat SM inhalational exposures while SM-induced leukopenia can be treated with granulocyte colony-stimulating factor (3). A specific antidote exists for lewisite that was secretly developed during World War II. British anti-lewisite (BAL, dimercaprol) will prevent or greatly decrease injury severity if applied within minutes of exposure. BAL binds to the arsenic in lewisite and can displace it from cellular components. Alternative heavy metal chelators can also be used as antidotes to include dimercaptopropane sulfonate and 2,3-dimercaptosuccinic acid (succimer). There is currently no antidote for phosgene oxime (2).

The Chemical Weapons Convention effectively banned the production, sale, or use of chemical weapons, established a destruction schedule for existing stockpiles, and created an enforceable inspection program (10). This treaty was ratified by the U.S. in 1997 and signatories include the vast majority of the United Nations members. Despite this international recognition of the threat and imposed disarmament some rogue nations continue to retain vesicant stockpiles. Terrorist groups have expressed an active interest in acquiring or developing chemical weapons like SM, which unfortunately is not difficult to produce. Because of these factors we must never forget the age old wisdom- a weapon once discovered never disappears... the vesicants will continue to be a formidable threat in the future.

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