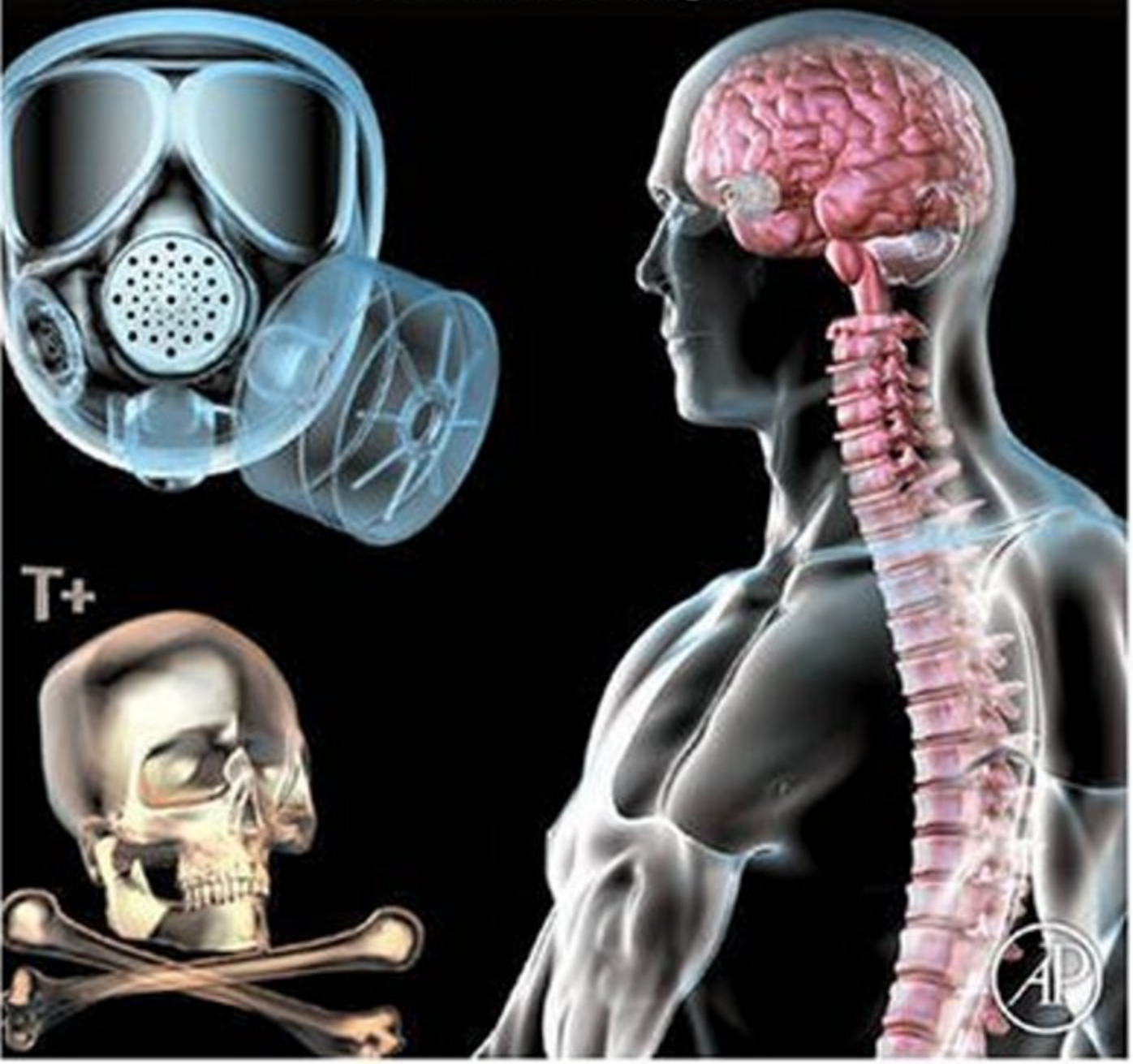
Handbook of Toxicology of Chemical Warfare Agents

EDITED BY

Ramesh C. Gupta



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This book is dedicated to my beloved wife Denise, daughter Rekha,
and parents the late Chandra and Triveni Gupta

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Foreword

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A highly toxic chemical intended to harm, kill, incapacitate, or control adversaries in warfare is called a chemical warfare agent (CWA). Throughout the world, some 70 different chemicals have been developed and produced, and many of them have been stockpiled as CWAs or chemical weapons of mass destructions (CWMD) during the 20th and 21st centuries. The use of biotoxins (botulinum toxin, ricin, saxitoxin, anthrax, etc.) is also sometimes referred to as chemical warfare.

Although the use of CWAs dates back to the 5th century BC, modern CWAs were not used at full scale until World War I. Since then, a variety of CWAs have been developed and deployed in many wars, conflicts, terrorist attacks, hostage crises, and riots. These chemicals came into the limelight particularly due to Gulf War Syndrome (aftermath of Gulf War I, 1991) and the Tokyo Subway terrorist attacks in 1994 and 1995. In the present world situation, the intentional use of highly toxic chemicals as CWAs/CWMD is a growing concern for government officials and civilians alike in developing as well as industrialized countries. The terrorist attacks in New York City on September 11, 2001 led to an increased awareness of protecting national monuments, landmarks, federal and state buildings, and workplaces, in addition to civilians, as possible terrorist targets.

Performing a comprehensive analysis of the inadequate database on chemical warfare agents is often a highly demanding task for toxicologists, risk assessors, regulatory agencies, and policy and decision makers at both state and federal levels. Due to the lack of adequate control, legislation, regulations, and knowledge, the term ''chemical warfare'' is often misunderstood or misused. In the recent past, in order to protect people and educate them about terrorist attacks, a variety of Standards and Exposure Guidelines have been made available, and on January 23, 2008, the U.S. Department of Labor published ''Safety and Health Topics, Chemical Terrorism''.

Handbook of Toxicology of Chemical Warfare Agents is the first book on chemical warfare agents that provides a plethora of knowledge on the historical perspective, epidemiology, detailed toxicology profile of CWAs/CWMD, target organ toxicity, analytical methodologies, biosensors, biomarkers, prophylactic and therapeutic countermeasures, and decontamination and detoxification procedures. In addition, the book serves as a significant source of information for nuclear and biological warfare agents. Therefore, this book appears to be an extremely useful reference source for academicians and regulatory authorities for risk and safety assessment, and management of chemical terrorism. The information provided in this book will draw immense attention from federal and state agencies, as well as political decision makers.

The editor, contributors, and publisher faced tremendous challenges in order to cover comprehensively all possible aspects of the toxicology of CWAs. Presently, there are at least two dozen chemicals that can be used as CWAs. Organophosphate (OP) nerve agents and mustards have been most frequently used and are most likely to be used in the future by terrorists and dictators throughout the world, because of their easy access and delivery systems. As a result, these chemicals have been extensively studied, and books, monographs, reviews, and papers are widely published. Recently, Academic Press/Elsevier published a most comprehensive book entitled *Toxicology of Organophosphate and Carbamate Compounds*, and *Handbook of Toxicology of Chemical Warfare Agents* appears to be unique in providing a thorough assessment of all possible aspects of toxicology, risk assessment, and remedial measures of CWAs in humans, animals, and wildlife.

The contributors of this book from around the globe are leading scientists and internationally recognized for their expertise in particular areas of toxicology of CWAs/CWMD and countermeasures. The book is divided into nine sections that deal with different aspects of CWAs. Section I deals extensively with the historical perspective, epidemiology and global impact of CWAs. Section II covers the broad array of chemical agents that can be weaponized and deployed as CWMD. In this section, toxicity profile, mechanism of action, risk assessment, and prophylactic/therapeutic measures of the individual chemicals are described in an in-depth manner. Section III provides an exhaustive coverage of target organ toxicity, which is indeed a novel aspect of this book. Several chapters on special topics about OP nerve agents, including molecular/

cellular mechanisms and neuropathological modulations, are discussed in section IV. Section V describes the risks to animals and wildlife associated with CWAs and chemicals of terror contaminating feed and water reservoirs, which can have a serious impact on human and animal health and the environment. Section VI deals with the metabolism, toxicokinetics and physiologically based pharmacokinetics of CWAs. A novel section (VII) is introduced with six chapters that provide discussion of analytical methodologies, biosensors, and biomarkers of CWAs. These topics will aid researchers in determining the extent of human/animal exposure, risk/safety assessment of CWAs, and management of poisoning. Section VIII covers extensively the unique approaches and strategies involved in prophylactic and therapeutic management and countermeasures. Many novel topics are included in this section, such as medical management of not only military personnel but civilians (more importantly the pediatric population), physiologically based pharmacokinetic modeling in countermeasures, catalytic and non-catalytic bioscavenging enzymes, and novel oximes. Prophylaxis and therapeutics for other CWAs are discussed in section II dealing with individual CWAs. The final section (IX) deals with information on decontamination and detoxification of CWAs.

In essence, this book is a landmark publication in the field of toxicology of CWAs/CWMD, as it provides comprehensive coverage of these chemicals and emphasizes current and novel issues that have not previously been addressed. It is hoped that this book will aid not only academicians but lay persons in community preparedness at local, state, and federal levels to protect civilians, military personnel, animals, wildlife, and the environment from chemical attacks by terrorists, dictators, and other adversaries. This book will be an invaluable source of information for homeland security, the Department of Defense, the Department of Veteran Affairs, the Department of Defense Research Establishment, diagnostic labs, poison control centers, federal, state and local authorities, forensic scientists, pharmacologists, toxicologists, chemists, biologists, environmentalists, teachers, students, and libraries.

CHAPTER 1

Introduction

RAMESH C. GUPTA

For centuries extremely toxic chemicals have been used in wars, conflicts, terrorists', extremists' and dictators' activities, malicious poisonings, and executions. One of the earliest forms of chemical warfare agents (CWAs) were natural toxins from plants or animals, which were used to coat arrowheads, commonly referred to as ''arrow poisons''. Ancient use of some CWAs and riot control agents (RCAs) dates back to the 5th century BC, during the Peloponnesian War, when the Spartans used smoke from burning coal, sulfur, and pitch to temporarily incapacitate and confuse occupants of Athenian strongholds. The Spartans also used bombs made of sulfur and pitch to overcome the enemy. The Romans used irritant clouds to drive out adversaries from hidden dwellings. In the 15th century, Leonardo da Vinci proposed the use of a powder of arsenic sulfide as a chemical weapon. Modern use of CWAs and RCAs or incapacitating agents dates back to World War I (WWI).

With advancements in science and chemistry in the 19th century, the possibility of chemical warfare increased tremendously. The first full-scale use of chemical warfare agents began in April of 1915 when German troops launched a poison gas attack at Ypres, Belgium, using 168 tons of chlorine gas, killing about 5,000 Allied (British, French, and Canadian) soldiers. During WWI, the deployment of CWAs, including toxic gases (chlorine, phosgene, cyanide, and mustard), irritants, and vesicants in massive quantities (about 125,000 tons), resulted in about 90,000 fatalities and 1.3 million non-fatal casualties. The majority of the deaths in WWI were a result of exposure to chlorine and phosgene gases. During the Holocaust, the Nazis used carbon monoxide and the insecticide Zyklon-B, containing hydrogen cyanide, to kill several million people in extermination camps. Poison gases were also used during the Warsaw Ghetto Uprising in 1943. Again, in November 1978, religious cult leader Jim Jones murdered over 900 men, women and children with cyanide.

Prior to, during, and after World War II, anticholinesterase organophosphate (OP) nerve agents/gases were developed in Germany, the USA, the UK, and Russia, and produced in large volumes in many other countries. They were maximally produced and stockpiled during the ''Cold War'' period. These nerve agents have been used in wars and by dictators, extremists, cult leaders, and terrorist groups as chemical weapons of mass destruction (CWMD) on many occasions. In 1980, Iraq attacked Iran, employing mustard and OP nerve gases. During the period of the Iraq and Iran conflict (1980–1988), Iran sustained 387 attacks and more than 100,000 troops were victims along with significant numbers of civilians. Thousands of victims still suffer from long-term health effects. Shortly after the end of the Iraq–Iran war in 1988, the brutal dictator of the Iraqi regime, Saddam Hussein, used multiple CWAs against the Kurdish minorities in Halbja village, killing more than 10% of the town's 50,000 residents. To date, mustards have been used in more than a dozen conflicts, killing and inflicting severe injuries in millions of military personnel and civilians.

During the Persian Gulf War, exposure to OP nerve agents occurred from the destruction of munitions containing 8.5 metric tons of sarin/cyclosarin housed in Bunker 73 at Khamisyah on March 4, 1991, and additional destruction of these nerve agents contained in rockets in a pit at Khamisyah on March 10, 1991. Although exposure levels to nerve agents were too low to produce signs of acute toxicity, the serving veterans in and around the Khamisyah area still suffer from long-term adverse health effects, most notably ''Gulf War Syndrome''. In 1996, about 60,000 veterans of the Persian Gulf War claimed to suffer from ''Gulf War Syndrome'' or ''Gulf Veterans' Illnesses'', possibly due to low-level exposure of nerve agents, mustard, pyridostigmine bromide and pesticides. Exposed veterans had an increased incidence of chronic myelocytic leukemia and increased risk of brain cancer deaths compared to unexposed personnel.

In the mid-1990s, two terrorist attacks by a fanatic religious cult Aum Shinrikyo (Supreme Truth), known as Aleph since 2000, took place in Japan (Matsumoto, 1994 and Tokyo Subway, 1995). In both incidents, the OP nerve agent sarin was used as a CWA. An estimated 70 tons of sarin was manufactured by Aum Shinrikyo in Kamikuishiki, Japan. Although the total fatality count involved not more than 20 civilians, injuries were observed in more than 6,000 and millions were terrified. These acts of chemical terrorism were unprecedented and the impact propagated not only throughout Japan, but the entire world. In the past few decades, many incidents have also occurred with biotoxins such as ricin and anthrax. Publicity surrounding frequent recent use due to easy access, and

copycat crimes increase the possibility of future use of these chemicals and biotoxins, which warrants advancement in emergency preparedness planning at the federal, state, and local government levels.

It is interesting to note that toxic chemicals have been used by governmental authorities against rebels, or civilians. In 1920s, Britain used chemical weapons in Iraq ''as an experiment'' against Kurdish rebels seeking independence. Winston Churchill strongly justified the use of ''poisoned gas against uncivilized tribes''. The Russian Osnaz Forces used an aerosol containing fentanyl anesthetic during the Moscow theater hostage crisis in 2002. RCAs were frequently used in the USA in the 1960s to disperse crowds in riot control.

At present, more than 25 countries and possibly many terrorist groups possess CWAs, while many other countries and terrorist groups are seeking to obtain them, due to their easy access. Some of these agents are stockpiled in enormous quantities and their destruction and remediation are not only expensive but associated with significant health risks. There is also the possibility of accidental release of CWAs or CWMD at the sites of their production, transportation, dissemination, and deployment. The intentional or accidental release of highly toxic chemicals, such as nerve agent VX (Dugway Proving Ground, Utah, 1968), Agent Orange (Vietnam, 1961–1972), PBB (Michigan, USA, 1973), dioxin (Seveso, Italy, 1976), and methyl isocyanate (Bhopal, India, 1984), has caused injuries in more than a million people, and deaths in several thousands. A 1968 accident with VX nerve gas killed more than 6,000 sheep in the Skull Valley area of Utah.

After September 11, 2001, the chances are greater than ever before for the use of CWMD by extremist and terrorist groups like Al Qaeda, which presents great risks to humans, domestic animals, and wildlife in many parts of the world. On 26 November 2008, Pakistani Islamic terrorists attacked Mumbai city in India at 10 different sites, including two luxury hotels, a Jewish center, a train station, and hospitals and cafes. Approximately 200 innocent people died and about 300 people were injured by bullets and fire smoke. It is more likely that these terrorist groups may use toxic industrial chemicals (agents of opportunity) either as such or as a precursor for more deadly CWMD. At present, many countries have established Defense Research Institutes with two major missions: (1) to understand the toxicity profile of CWAs/CWMDs, and (2) to develop strategic plans for prophylactic and therapeutic countermeasures. By the turn of the 21st century, the USA established the Department of Homeland Security. Many other countries also developed similar governing branches and agencies at the state and national level to protect people and properties from terrorist attacks. Among chemical, biological, and radiological weapons, the possibility of CWMD is more likely because of their easy access and delivery system. It is important to mention that understanding the toxicity profile of CWAs/CWMD is very complex, as these chemical compounds are of a diverse nature, and as a result, treatment becomes very difficult or in some cases impossible.

In the past, many accords, agreements, declarations, documents, protocols, and treaties have been signed at the international level to prohibit the development, production, stockpiling, and use of CWAs, yet dictators and terrorists produce and/or procure these chemicals to harm or kill enemies, create havoc, and draw national and international attention. In 1907, The Hague Convention outlawed the use of chemical weapons, yet during WWI, many countries used these chemicals. The first international accord on the banning of chemical warfare was agreed upon in Geneva in 1925. Despite the General Protocol, the Japanese used chemical warfare against China in 1930. In 1933, the Chemical Weapon Convention banned the development, possession, and use of CWAs. The document was signed and implemented by more than 100 countries. Yet, during WWI many chemicals of warfare were developed, produced, and used by many countries. In 1993, another global convention banning the production and stockpiling of chemical warfare agents was signed by more than 100 countries.

In the present world situation, it is highly likely that these agents will be used in wars, conflicts, terrorist attacks, and with malicious intent. In such scenarios, these extremely toxic agents continuously pose serious threats to humans, animals, and wildlife.

This *Handbook of Toxicology of Chemical Warfare Agents* was prepared in order to offer the most comprehensive coverage of every aspect of the deadly toxic chemicals that can be used as CWAs/CWMD. In addition to the chapters on radiation, several chapters are included on deadly biotoxins (ricin, abrin, strychnine, anthrax, and botulinum toxins) that can be weaponized in chemical, radiological, and biological warfares. Many special and unique topics are offered that have not been covered in previous books. This is the first book that offers detailed target organ toxicity in this area of toxicology. In every chapter, all factual statements are substantiated with appropriate references.

This book meets the needs not only of academicians but lay persons as well. The format employed is user friendly and easy to understand. Standalone chapters on individual chemicals, target organ toxicity, biosensors and biomarkers, risks to man, animal and wildlife, and prophylactic and therapeutic countermeasures are just a few of the many novel topics covered in this book. The chapters are enriched with the historical background as well as the latest information and up-to-date references. With more than 70 chapters, this book will serve as a reference source for toxicologists, pharmacologists, forensic scientists, analytical chemists, local/state/federal officials in the Department of Homeland Security, Department of Defense, Defense Research Establishments, Department of Veterans Affairs, physicians at medical and veterinary emergency care units of hospitals, poison control centers, medical and veterinary diagnostic

labs, environmentalists and wildlife interest groups, researchers in the area of nuclear, chemical, and biological warfare agents, and college and university libraries.

Contributors of the chapters in this book are the most qualified scientists in their particular areas of chemical and biological warfare agents. These scientists are from around the globe and regarded as authorities in the field of pharmacology, toxicology, and military medicine. The editor sincerely appreciates each author for his/her dedicated hard work and invaluable contributions to this volume. The editor gratefully acknowledges Robin B. Doss and Kristie A. Rohde for their technical assistance, Alexandre M. Katos for cover design and Denise M. Gupta for indexing. Finally, the editor remains indebted to Renske Van Dijk, Rebecca Garay, and William Brottmiller, the editors at Elsevier, for their immense contributions to this book.

CHAPTER 2

Historical Perspective of Chemical Warfare Agents

NATHAN H. JOHNSON, JOSEPH C. LARSEN, AND EDWARD MEEK

The opinions or assertions contained herein are the private views of the authors, and are not to be construed as reflecting the views of the Department of Defense, the Defense Threat Reduction Agency, or the US Air Force.

I. INTRODUCTION

The employment of chemicals in warfighting has a long history (Silvagni *et al.*, 2002; Romano *et al.*, 2008). Just as the utilization of chemicals brought about tremendous advances in society, the concept of using chemicals as a contributing factor in winning wars has been pursued for centuries (Joy, 1997; Smart, 1997). There are many examples of the exploitation of chemicals in warfare and conflict dating back to antiquity. Primitive man may have been the first to use chemical compounds in hunting and in battle. The use of smoke from fires to drive animals or adversaries from caves may have been the earliest use of chemical weapons. Natural compounds from plants, insects, or animals that were observed to cause sickness or death were likely used by our distant forefathers in attempts to gain or maintain superiority (Hammond, 1994). Natural toxins from plants or animals on arrowheads or the poisoning of water or food could increase casualties and cause fear in opposing military forces or civilian populations. These early uses of chemicals would pave the way for more lethal chemical weapons. For example, in the fourth century BC, smoke containing sulfur was used in the war between Sparta and Athens (Joy, 1997). Chinese manuscripts indicate arsenical-based compounds were used in conflict (Joy, 1997). A few hundred years later, toxic smoke was used by the Romans in Spain (Coleman, 2005). During the second siege of Constantinople, the Byzantine emperor Leo III used ''Greek fire'' in his quest for military victory (Coleman, 2005). During the ensuing years, there were many instances of the limited and attempted use of chemicals and toxins on the battlefield. Many of these examples may have been influenced by the intentional poisonings occurring in civilian settings (Joy 1997; Smart, 1997; Newmark, 2004; Coleman, 2005). The earliest known treaty to ban poisons in warfare was signed between the French and Germans in the 17th century (Smart, 1997). In the siege of Groningen, incendiary devices were used by European armies to release belladonna, sulfur, and other compounds. This led to the Strasbourg Agreement in 1675 (Coleman, 2005). This agreement prohibited poison bullets (Smart, 1997).

As science and chemistry advanced in the 19th century, the possibilities of chemical warfare increased exponentially. Advancements were made in industrial applications of sulfur, cyanide, and chorine (Joy, 1997). In addition, the concept of chemicals in projectiles was introduced. During the Crimean War, the British refused to use cyanide-based artillery shells against the Russians on the grounds that it was a ''bad mode of warfare'' (Smart, 1997). This was an early example of the ethical questions surrounding chemical use in warfare that continued into the 20th century (Vedder and Walton, 1925). During the American Civil War, both the Northern and Southern armies seriously considered using various chemicals in their pursuit of operational victories (Smart, 1997). Early attempts at international treaties were met with mixed results. The USA prohibited any use of poison in the Civil War. The Brussels Convention on the Law and Customs of War of 1874 prohibited poisons or poison-related arms (Smart, 1997). The first Peace Conference at The Hague prohibited projectiles filled with asphyxiating or deleterious gases (Smart, 1997). Some countries, including the USA, were not signatories to this agreement. The employment of chemicals as asphyxiating warfare agents was vigorously discussed at The Hague convention (Joy, 1997). Arguments were again made against chemicals based on moral grounds. However, counterarguments were made based on the assumption that chemicals lead to a death devoid of suffering (Vedder and Walton, 1925; Joy, 1997; Coleman, 2005). Individuals who advocated chemicals did not see their use as an unfair advantage; rather, it was just one in a series of technological advances, which if mastered could provide strategic, operational, and tactical advantages on the battlefield. The second Peace Conference at The Hague 8 years later prohibited poisons or poisoned weapons (Smart, 1997). The British use of picric acid-filled shells during the Boer War and the Japanese use

of arsenical rag torches in the Russo-Japanese War further illustrate that chemical warfare was considered by some a legitimate form of warfare at the turn of the 20th century (Smart, 1997). During the early 20th century, technological advancements in the chemical industry made the possibility of sustained military operations using chemicals a realistic possibility. The murder of Archduke Francis Ferdinand at Sarajevo set the stage for what would become the first widespread use of chemical weapons to date (Harris and Paxman, 2002).

II. THE FIRST SUSTAINED USE OF CHEMICALS AS AGENTS OF WARFARE

The talk and rhetoric of the late 19th century should have prepared the countries involved in World War I for chemical warfare. However, that was not case (Smart, 1997). World War I clearly demonstrated the deadly and destructive nature of chemicals in modern warfare. Both alliances in the war experimented with novel forms of warfare, to include chemical weapons, and followed the lead of their advisory (Hay, 2000). It is little wonder this war is known as the ''chemist's war'' (Fitzgerald, 2008). Initially, the French used gas grenades with little effect and were followed by the German use of shells filled with tear gas (Joy, 1997). The Germans, capitalizing on their robust chemical industry, produced shells filled with dianisidine chlorosulfate (Smart, 1997). These shells were used in October of 1914 against the British at Neuve-Chapelle but had little effect. In the winter of 1914–15, the Germans fired 150 mm howitzer shells filled with xylyl bromide (Smart, 1997). The xylyl bromide shells were fired on both the eastern and western fronts with disappointing effects. Despite the inauspicious start of chemical warfare on both fronts, efforts were continued to develop new uses. It would soon be evident that chemical warfare would be devastating on the battlefield (Coleman, 2005; Tucker, 2006). Fritz Haber, a German scientist who later won the Nobel prize in Chemistry, had proposed the possibility of releasing chlorine gas from cylinders (Joy, 1997). Chemical warfare was attractive to Germans for two reasons: the shortage of German artillery shells and the ability to defeat the enemy trench system (Smart, 1997). After consideration and debate, the Germans released chlorine in April 1915 at Ypres, Belgium (Coleman, 2005). The German military was not prepared for the tremendous operational advantage the chlorine release provided. It did not take long for the British and French forces to respond in kind to the German offensive (Vedder and Walton, 1925; Joy, 1997; Smart, 1997; Coleman, 2005). In the fall of 1915, a British officer, William Livens, introduced a modified mortar (Figure 2.1) that could project gas-filled shells of chlorine or phosgene, the two agents of choice at that time (Joy, 1997). Both chlorine and phosgene caused extreme respiratory problems to those soldiers who were exposed (Vedder and Walton, 1925; Joy, 1997; Smart, 1997; Coleman, 2005; Hurst *et al.*, 2007) (Figure 2.2).

As the USA entered the war in the spring of 1917, an obvious concern of the military command was the effect of chemical warfare on standard operations. Chemistry departments at universities were tasked with investigating and developing novel chemical agents (Joy, 1997). Protective equipment (Figure 2.3) and basic studies of the biological effects of chemical agents were assigned to the US Army Medical Department (Joy, 1997). In the fall of 1917, the Army began to build an industrial base for producing

FIGURE 2.1. British Livens Projector, Western Front, World War I.
Source: United Kingdom Government (http://en.wikipedia.org/wiki/Image:Livens_gas_projector_loading.jpg)

FIGURE 2.2. Australian infantry in trench with gas masks donned, Ypres, Belgium, September 1917. Photo by Captain Frank Hurley (http://en.wikipedia.org/wiki/Image:Australian_infantry_small_box_respirators_Ypres_1917.jpg)

FIGURE 2.3. US Army captain wearing a gas mask in training, 1917.
Source: Library of Congress.

chemical agents at Edgewood Arsenal, Maryland (Joy, 1997). As the effects of chlorine and phosgene became diminished by the advent of gas masks (Figure 2.4), the Germans turned to dichlorethyl sulfide (mustard) at Ypres against the British (Joy, 1997). As opposed to the gases, mustard remained persistent in the area and contact avoidance was the major concern (Joy, 1997). It is worth noting that almost 100 years after it was first used on the battlefield, mustard still has no effective treatment and research continues for effective therapeutics (Babin and Ricketts *et al.*, 2000; Baskin and Prabhaharan, 2000; Casillas and Kiser, 2000; Hay, 2000; Schlager and Hart, 2000; Hurst *et al.*, 2007; Romano *et al.*, 2008). It has been estimated that there were over one million chemical casualties (Figure 2.5) of World War I with almost 8% being fatal (Joy, 1997). The Russians on the eastern front had a higher percentage of fatalities when compared with other countries in the war, primarily due to the later introduction of a protective mask (Joy, 1997). The relatively low mortality rate of chemical casualties in World War I demonstrated the most insidious aspect of their use, the medical and logistical burden it placed on the affected army. The eventual Allied victory brought a temporary end to chemical warfare. In 1919, the Treaty of Versailles prohibited the Germans from production and use of chemical weapons.

III. INITIAL COUNTERMEASURES

The conceptualization of a protective mask dates back over 500 years to Leonardo da Vinci (Smart, 1997). By the mid-19th century, protective masks were proposed in the USA and Europe for both industrial and military use. The modern ''gas mask'' was developed by the Germans with sodium thiosulfate and bicarbonate soaked pads and used in World War I (Joy, 1997). The French and English soon followed with their own versions of gas masks (Joy, 1997). In 1916, the Germans introduced a mask that incorporated a canister through which the soldiers breathed (Joy, 1997). Initially, the American forces in World War I used gas masks obtained from allies already fighting in the war (Smart, 1997). In 1918, the Americans introduced their RFK mask, a modified version of the British mask. Masks were also developed for the animals that supported the war fighting efforts. Decontamination efforts during World War I were rudimentary and included chemical neutralization and aeration of clothing and equipment. Although the need for

FIGURE 2.4. World War I soldier and horse wearing gas mask. *Source*: National Archives and Records Administration (http://commons.wikimedia.org/wiki/Image:Gasmask_for_man_and_horse.jpeg)

detection of chemical agents was clearly identified, very little progress was made during World War I. Medical treatment included removal of the patient from the source, decontamination, and palliative care (Smart, 1997).

IV. EVENTS AFTER WORLD WAR I

At the conclusion of World War I, the world had been introduced to chemical warfare on an unprecedented level. While there were groups that thought that humanity had learned a lesson from the cruel nature of chemical warfare, others prudently went to work on improved chemical defense (Vedder and Walton, 1925). The thoughts of many professional military officers were that future wars would be fought under the new paradigm of chemical warfare (Vedder and Walton, 1925; Vedder, 1926; Smart, 1997). New gas masks were developed and training in chemical environments was introduced (Vedder and Walton, 1925; Vedder, 1926; Joy, 1997). Textbooks and manuals, such as those written by US Army Colonel Edward B. Vedder (Figure 2.6), were introduced to the military medical community (Vedder and Walton, 1925). In addition, the civilian medical community gained valuable insight into toxicology and animal models from the events of World War I (Vedder, 1929; Johnson, 2007). Despite the first-hand experience with chemical warfare, some countries, including the USA, struggled to adequately fund their offensive and defensive programs during demobilization (Smart, 1997). It did not take long for chemical warfare to appear in other conflicts. Chemical agents were used to subdue rioters and suppress rebellions. The British used chemical agents to suppress uprisings in Mesopotamia by dropping bombs in cities throughout the area in the early 1920s (Coleman, 2005). The Soviet Union used chemical agents to quell the Tambov rebellion in 1921, and France and Spain used mustard gas bombs to subdue the Berber rebellion during the 1920s (Werth *et al.*, 1999). Italy and Japan used mustard in small regional conflicts (Joy, 1997). The Italian conflict in Ethiopia was noteworthy because

FIGURE 2.5. British soldiers temporarily blinded by tear gas awaiting treatment at the Battle of Estaires, April 1918. Photo by 2nd Lt T.L. Aitken.
Source: United Kingdom Government (http://en.wikipedia.org/wiki/Image:British_55th_Division_gas_casualties_10_April_1918.jpg)

FIGURE 2.6. Captain Edward Vedder, ''the father'' of USAMRICD. Photo courtesy of Mrs Martha Vedder.

mustard was sprayed and dropped from planes and the agent's use was considered by some to be significant to the Italian victory (Smart, 1997). This use demonstrated the contemporary thought that allowed chemicals to be viable alternatives to traditional combat. The Japanese also used chemical weapons during the 1930s against regional foes. Mustard gas and the vesicant Lewisite were released on Chinese troops and were also used in South East Asia (Coleman, 2005). Lewisite is an arsine which was usually produced as an oily brown liquid that was said to have the odor of geraniums (Spiers, 1986; Hammond, 1994). It was developed in the USA by Winford Lee Lewis in 1918 and was found to be effective at penetrating clothing. The USA produced approximately 20,000 tons of Lewisite but only used small quantities of the chemical in World War I (Coleman, 2005). Dimercaprol, more commonly called British anti-Lewisite, was developed as an effective treatment for the vesicant (Goebel, 2008). In the inter-war period, mustard was a key concern in defensive planning (Coleman, 2005). New stores of mustard were produced in many countries. Work continued on many fronts to improve protective equipment. For example, the US Chemical Warfare Service introduced the M1A2 mask, an improvement of the M1 mask (Smart, 1997). In the Geneva Protocol of 1925, 16 of the world's major nations pledged never to use gas as a weapon of warfare; it was not ratified in the USA until 1975 (Hammond, 1994). There has long been vigorous debate on the merits of treaties with nations balancing the military needs versus the potential irrational concept of chemical warfare (Vedder, 1926).

V. WORLD WAR II

In the lead up to World War II, the Germans forever changed chemical warfare with the discovery of the organophosphorus nerve agents (Goebel, 2008). These organophosphorus-containing nerve agents inhibit cholinesterase enzyme in the nerve synapse responsible for the breakdown of the neurotransmitter acetylcholine (ATSDR, 2008). This results in the accumulation of the neurotransmitter in the synapse and overstimulation of the nervous system. This can result in subsequent respiratory failure and death (ATSDR, 2008).

In 1936, Gerhard Schrader, a German chemist working on the development of insecticides for IG Farben, developed a highly toxic organophosphate compound which he named ''tabun'' (Hersh, 1968; Hammond, 1994). Schrader and an assistant became a casualty of their discovery when a drop of the neurotoxicant was spilled in the lab exposing both of them (Tucker, 2006). Had the amount of tabun spilled been greater both researchers would have certainly succumbed to the effects of the poison. Tabun was the first member in a series of compounds termed ''nerve gases'' (Coleman, 2005). The correct terminology is ''nerve agents'' as these agents are not gases, but liquids dispersed as fine aerosols. Tabun was extremely toxic in small amounts and invisible and virtually odorless (Tucker, 2006). The compound could be inhaled or absorbed through the skin. These characteristics made it too dangerous to be used as an insecticide by farmers. German law required that any discovery having military application be reported to military officials (Tucker, 2006). Schrader was not overly excited about producing chemical agents for the military; however, the Germans placed him in a secret military research facility with the emphasis on producing these nerve agents and discovering new agents (Tucker, 2006). Subsequently, Schrader and his team of researchers discovered a more lethal organophosphate compound similar to tabun, which he named ''sarin'' in honor of the team members: Schrader, Ambrose, Rudriger, and van der Linde (Coleman, 2005).

At the onset of World War II, both the Allies and the Germans anticipated chemical agents would be deployed on the battlefield (Tucker, 2006). This expectation intensified research into the development of new agents, delivery systems, and methods of protection (Figures 2.7 and 2.8). The Allied forces were unaware of the Germans' new nerve agent, tabun, at the beginning of the war. The rapidly advancing German army offered very little opportunity to use chemical agents, as it could prevent the rapid movement of the German troops into an area after being released (Tucker, 2006). Nevertheless, the Germans produced and stockpiled large amounts of nerve agents throughout the war (Spiers, 1986). The production of these organophosphate agents was complex, required custom equipment, and was hazardous to those involved in production (Tucker, 2006). If exposed, the workers would be dunked in a bath of sodium bicarbonate (Harris and Paxman, 2002; Goebel, 2008). It is also interesting to note that some members of the German workforce were given rations containing higher percentages of fat (Harris and Paxman, 2002). This was done because

FIGURE 2.7. Gas mask production – Detroit, Michigan, 1942. *Source*: Library of Congress.

FIGURE 2.8. World War II: a private trains using protective gear. Photo courtesy of the US Army Medical Research Institute of Chemical Defense.

authorities observed that workers with higher quality rations seemed protected against exposure to low levels of tabun. Many detainees were used in the manufacture and testing of chemical agents in Germany (Harris and Paxman, 2002; Tucker, 2006). It is not known how many chemical casualties there were in prisoners of war due to their forced labor in nerve agent production, but documented fatalities were recorded. The discovery of tabun and sarin was followed by the discovery of soman by Richard Kuhn and Konrad Henkel at the Kaiser Wilhelm Institute for Medical Research in 1944 (Tucker, 2006). This class of nerve agents is collectively termed ''G'' agents; the G is for German, since German researchers discovered this class of compounds. A second letter is included as the specific identifier of each compound: GA (tabun), GB (sarin), GD (soman), and GF (cyclosarin) (ATSDR, 2008). These agents were mass produced by the Nazi regime throughout the war but were not used (Tucker, 2006). There has been considerable debate questioning why the Germans did not employ their chemical weapons in World War II. While it may never be conclusively known, several potential reasons include a lack of intelligence regarding the German superiority in chemical weapons discovery, fear of retaliation, and Adolph Hitler's personal exposure to chemical agents on the battlefield in World War I (Harris and Paxman, 2002; Tucker, 2006).

Other chemical agents that had been produced during and following World War I were still being produced. On December 2, 1943, German planes sank several American ships off the coast of Italy and at least one of the ships contained mustard that was to be used as a retaliatory response if the Germans unleashed a large-scale chemical weapons attack (Tucker, 2006). Casualties resulted from exposure to the mustard, some of which were inflicted on civilian merchant seamen (US Navy, 2008). The presence of the agent on the ship was classified and resulted in incorrect treatment of many of the exposed by physicians (Tucker, 2006).

VI. POST-WORLD WAR II

By the conclusion of World War II, both the Allies and Germany had stockpiled large amounts of chemical agents (Tucker, 2006). The Allied forces divided up the stockpiles of agents discovered in German facilities. Following the end of the war, many of the Allied countries continued to conduct research on the German nerve agents. The rise of the Soviet Union as a power and adversary prompted the USA and other countries to continually search for novel chemical and biological warfare agents (Tucker, 2006). The research and resources that were allotted for these efforts were not trivial even though they were often overshadowed by the research and development of thermonuclear weapons (Hersh, 1968; Goebel, 2008).

The post-World War II era ushered in the nuclear age. Some felt the age of chemical warfare was past (Smart, 1997). Events would prove this to be a hasty conclusion. In the USA, research of the G-series agents and medical countermeasures against these agents was accomplished by

the late 1940s. Research and intelligence gathering was further hastened by the impressive gains the Soviet Union made in chemical warfare capability in the years after World War II. By the early 1950s, production of sarin had been initiated in the USA (Smart, 1997). At nearly the same time, Ranajit Ghosh, a chemist at the British Imperial Chemical Industries plant, developed a new organophosphate compound as a potential insecticide (Tucker, 2006). Like Gerhard Schrader before him, this compound was deemed too toxic to be used in the field as a pesticide. The compound was sent to researchers in Porton Down, England, synthesized and developed into the first of a new class of nerve agents, the ''V'' agents (Goebel, 2008). Like the ''G'' agents the ''V'' agents also have a second letter designation: VE, VG, VM, and VX (Coleman, 2005). Of these agents, VX was the most commonly produced. The ''V'' series of agents are generally more toxic than the ''G'' agents (ATSDR, 2008). In a deal brokered between the British and US governments, the British traded the VX technology for thermonuclear weapons technology of the USA (Tucker, 2006). The USA produced and stockpiled large quantities of VX (Hersh, 1968; Hammond, 1994).

Throughout the 1950s and 1960s, advancements were made in production and delivery of chemical weapons to include sarin and VX (Smart, 1997). While work on improved masks continued, a renewed concern was the inability to detect nerve agents. Several prototypes were developed in the mid-1950s. Great advancements were made in therapeutics of agents that inhibited the enzyme acetylcholinesterase (Gupta, 2006; Taylor, 2006; Klaassen, 2008). Atropine was introduced in the early 1950s. Oximes were added as an adjunct to speed up reactivation of the enzyme (Smart, 1997). The autoinjector was developed to overcome user fear of self-injection of atropine. Major advances were made in utilization of chemical weapons in artillery (Figure 2.9). For example, the USA developed both short and long range rockets filled with chemical agent. The USA disposed of stockpiles of its chemical weapons in the late 1960s in an operation termed CHASE (cut holes and sink em) in the sea (Coleman, 2005). In 1969, nerve agent stockpiles were discovered in US depots in Japan after several US military servicemen became ill while doing maintenance (Tucker, 2006). This stockpile had been kept secret from the Japanese and created an uproar that resulted in the later disposal of the agents in the Johnston Atoll in the Pacific Ocean.

Defensive equipment such as improved field alarms and drinking tubes for gas masks were introduced in the 1960s (Smart, 1997). Great strides were also made in collective protection in the 1960s and 1970s. Although not used extensively since World War I, chemical agents have nonetheless been used for military purposes. The Egyptians allegedly used mustard and possibly nerve agents in the Yeman civil war (Joy, 1997; Smart, 1997). This was the first reported use of nerve agent in armed conflict. There were allegations that chemical agents were used by the Vietnamese in Laos and Kampuchea in the late 1970s (Coleman, 2005). In the Vietnam War, the USA used defoliants and tear gas (Joy, 1997). The Soviet Union was accused of using chemical agents in their war in Afghanistan (Joy, 1997).

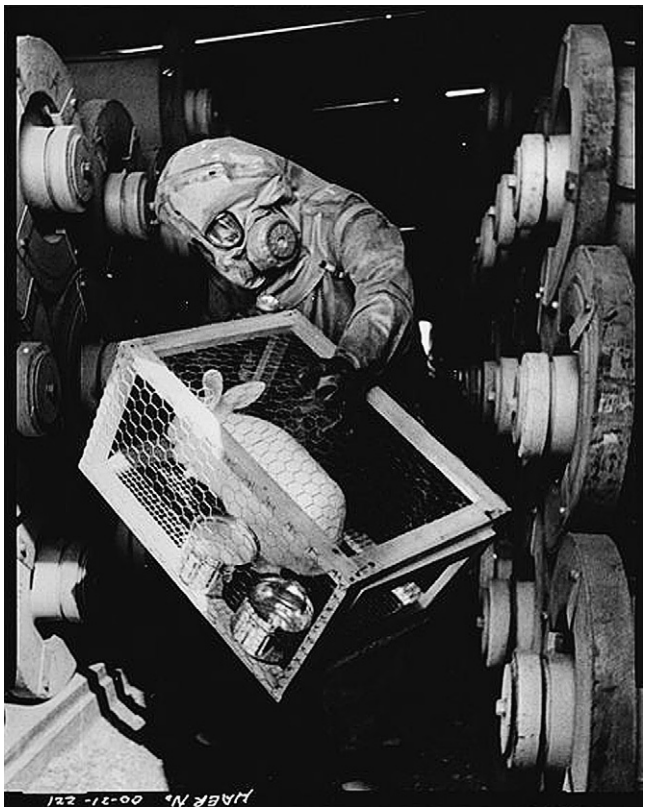

FIGURE 2.9. Testing for leaks at Sarin production plant, 1970. *Source*: Library of Congress (http://memory.loc.gov/pnp/habshaer/co/co0100/co0168/photos/316333pr.jpg).

VII. INCAPACITANTS AND TOXINS

Incapacitating agents have long been considered an intermediate between chemical and traditional warfare. The Germans investigated the military use of lacrimators in the 1880s followed shortly thereafter by the French (Smart, 1997). The English and French considered using lacrimators in World War I (Smart 1997). Japanese forces used tear gas against the Chinese in the late 1930s. The US Army used riot control agents and defoliants in the Vietnam War (Smart, 1997). The defoliant ''Agent Orange'' was later potentially linked to several forms of cancer (Stone, 2007). During the 1950s and 1960s, the USA had an active incapacitant program (Smart, 1997). These agents were thought of as more humane than traditional chemical agents because the intent was not lethality. These agents were designated ''K-agents'' and included tetrahydrocannabinol and lysergic

acid (Smart, 1997). One of the most extensively studied incapacitating agents was 3-quinuclidinyl benzilate, designated BZ by the US Army (Ketchum, 2006). Like many incapacitating agents, BZ was not adopted due to difficulty producing reproducible effects, unwanted side effects, latency to produce effects, and difficulty in producing a dissemination that was free of smoke (Smart, 1997; Ketchum, 2006).

There have been multiple attempts to use the toxins from plants and living organisms to develop viable weapon systems. Two that are noteworthy are ricin and botulinum toxin. Ricin has been recognized as a potential biological weapon since World War I. While the British were developing the V agents, US military researchers patented a procedure for purifying ricin, a very potent toxin from the castor bean plant (Harris and Paxman, 2002). The development of a method of dissemination of ricin as a chemical weapon proved problematic thus making its use very limited. In 2003, ricin was detected on an envelope processed in a Greenville, South Carolina, postal facility. Postal workers did not develop symptoms of ricin exposure and the individual who mailed the letter remains at large (Shea, 2004). The development and use of botulinum neurotoxin as a biological weapon was initiated at least 60 years ago (Smart, 1997; Arnon, 2001). In the 1930s, during occupation of Manchuria, the Japanese biological warfare group, Unit 731, purportedly fed cultures of *Clostridium botulinum* to prisoners causing human lethality. The US Army biological weapons program produced botulinum neurotoxin during World War II in response to Germany's biological weapons program (Coleman, 2005). In fact, more than 100 million toxoid vaccine doses were prepared and forward positioned in time for the D-Day invasion of Normandy (Arnon, 2001).

VIII. RECENT EXPERIENCES

The 1980s proved to be very significant in the employment of chemical weapons on the battlefield. In 1980, Iraq invaded Iran (Smart, 1997). The Iraqi armed forces, who were advised by the Soviet Union, possessed chemical agents and were trained in their use. The war was unequivocally barbarous and neither side gained an advantage. In many ways, this war had similarities to World War I. By 1983, Iran formally protested to the United Nations about the Iraqi use of chemical agents. The general consensus was that Iraq used mustard agent and possibly tabun in this war (Figure 2.10). It is estimated that 5% of Iranian casualties, totaling approximately 45,000, can be attributed to chemical warfare agents (Smart, 1997). The same author also reported that the Iraqi Army used chemical agents against the Kurdish minority in northern Iraq. Lybia was also suspected of using chemical agents when Chad was invaded in 1986 (Smart, 1997).

FIGURE 2.10. Aftermath of Iraqi chemical weapon attack (1980s). Photo by Sayeed Janbozorgi; image used under the terms of the GNU free documentation license (http://en.wikipedia.org/wiki/Image:Chemical_weapon2.jpg).

The late 1980s also saw improvements in defensive equipment such as the M40 gas mask developed by the USA (Smart, 1997). Other advancements were made in collective protection, decontamination, and detection. In 1984, US President Ronald Reagan issued a statement calling for an international ban on chemical weapons (Tucker, 2006). Subsequently, on June 1, 1990, President George H.W. Bush and Soviet Union leader Mikhail Gorbachev signed a treaty banning the production of chemical weapons and initiated the destruction of the stockpiles of both nations (Tucker, 2006). In 1993, the Chemical Weapons Convention was convened and signed. It was implemented in 1997 (Hammond, 1994). As of 2008, the vast majority of United Nations member states have joined the Chemical Weapons Convention (OPCW, 2008).

In 1990, the Iraqi Army invaded neighboring Kuwait. Subsequently, the USA and eventually a coalition sent forces to the area at the request of Saudi Arabia (Smart, 1997). Because of the broad knowledge of Iraqi chemical use on the battlefield in the 1980s, coalition forces were the largest force to operate in a potential chemical environment since World War I. Forces moved into the area of operation were provided with atropine autoinjectors, an acetylcholinesterase reactivator, and a nerve agent pretreatment (pyridostigmine bromide). Fortunately, chemical weapons were not apparently used in this conflict, although multiple false alarms were reported. The failure of the Iraqi military to use chemical weapons could be attributed to fear of retaliation, breakdown of communication, changing wind patterns, the surprising speed of the coalition attack, or the fact that Iraqi chemical infrastructure was attacked during the initial portion of the conflict. There have been many coalition veterans who report a myriad of symptoms that

have been commonly referred to as ''Gulf War Syndrome''. The etiology of this syndrome is unclear despite multiple epidemiological studies (Coleman, 2005).

IX. TERRORIST USE

One of the reasons why chemical weapons have been used relatively infrequently in combat over the past century is the fear of retaliation by opposing countries. In less organized asymmetrical conflicts, the fear of retaliation is of less concern. The potential exploitation of chemical weapons by terrorists is of great worldwide concern. The appeal of these weapons to terrorists is centered on the fact that many of the chemical agents are cheap and relatively easy to produce, transport, and release. These characteristics, along with the fear associated with the idea of a chemical attack, make chemicals an ideal weapon for creating terror (Romano and King, 2001). In 1974, Muharem Kurbegovic attacked several public buildings with firebombs in California and claimed to have developed sarin and some other nerve agents (Tucker, 2006). The search of his home resulted in the discovery of various precursor materials for chemical agents and a large amount of sodium cyanide. In 1994, the Aum Shinrikyo, a Japanese religious cult, carried out several attacks using sarin produced by the cult's members (Tucker, 2006). The attacks included a residential and subway exposure. A total of 19 people were killed and over 6,000 sought medical attention. Some of those seeking medical attention may be attributed to a fear of exposure. Psychological stress is a common aftermath of a chemical or biological attack (Romano and King, 2001). In the 21st century, formerly used chemicals of military interest have reemerged as contemporary threats. In the fall of 2006, Al Qaeda and associated groups used chlorine combined with traditional car and truck bombings to spread panic in Iraq (Garamone, 2007). These attacks were followed by similar attacks in the subsequent months.

X. CONCLUDING REMARKS AND FUTURE DIRECTION

As long as there are legitimate uses for chemicals in our society, the risk of chemical agents in conflict and terrorist activity will always be present. Research across the globe continues for better detection, protection, and treatment of chemical warfare agents. While many countries have denounced and are signatories to various treaties to limit the use and production of chemical warfare agents, non-state and terror organizations are under no such restrictions. Luckily, chemical weapon use has been limited in warfare and conflict. As we progress into the 21st century, the use of established chemical warfare agents is a real possibility. The potential use of legitimate industrial chemicals (e.g. the Iraqi burning of petroleum fields in the first Gulf War) and the potential synthesis of new agents should also be recognized. History has demonstrated that chemicals have been used in both organized and asymmetrical conflicts and preparations for defense and therapy for such encounters is prudent. Chemicals represent a unique force multiplier that simply cannot be ignored in the 21st century.

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CHAPTER 3

Global Impact of Chemical Warfare Agents Used Before and After 1945

JIRI BAJGAR, JOSEF FUSEK, JIRI KASSA, KAMIL KUCA, AND DANIEL JUN

I. INTRODUCTION

The threat of chemical weapons (CWs), used either by States or Parties to the Chemical Weapons Convention (Convention on the Prohibition of the Development, Production, Stockpiling and Use of Chemical Weapons and on their Destruction) or by terrorists, has never attracted so much public attention as in the past 10 years. In spite of the existing legal documents dealing with prohibition of CWs, e.g. Geneva Protocol 1925, and Chemical Weapons Convention (CWC), some incidents of the use of CWs in different conflicts and terroristic attacks have been observed. Moreover, alleged use of CWs was noted during the period from 1925 to the present time. It must be emphasized that the theoretical and practical basis for production, storage, and employment of CWs still exists. Also, it must be clearly stated that CWs are applicable at any time, in any place, and in large quantities.

CWs consist of the chemical warfare agents (CWAs) and the means to deliver to the target. They are characterized by high effectivity and large targets and are known as area weapons or silent weapons. They are relatively low cost and with their use it is possible to achieve destruction of everything that is living but avoid destruction of materials and buildings. They are also called the nuclear weapons of poor countries – ''poor man's nuclear weapon''. It should be pointed out that the use of CWs is connected with the use or release of toxic chemicals, thus, chemical warfare can be considered part of generally observed situations where toxic chemicals are used or released and influence the environment and humankind.

There exist a number of causal reasons for these events but apart from accidents connected with the release of toxic chemicals from a natural source (e.g. volcanoes), the factors shown in Figure 3.1 or their combinations can be involved.

For military purposes, a number of chemicals were tested, but only a small number are contained in military arsenals. However, according to the definition contained in the CWC, any toxic chemical intended for military use must be considered a chemical weapon, i.e. the aim is to limit the designation of the compound in question for use as a CW. On the other hand, all toxic chemicals of high toxicity can be chosen by terrorists.

II. BACKGROUND

The use of toxic chemicals against humankind is as old as any warfare conflict. The use of the poisoned arrow against man – not animal – can be considered as the beginning of chemical warfare and would be characterized as the intentional use of chemicals.

At the very beginning, chemical warfare was more closely connected with fire. ''Greek fire'' was an excellent naval weapon because it would float on water and set fire to the wooden ships. There are other examples from history: for example, toxic smoke was used in in China in 2000 BC. In Thucydides' *History of the Peloponnesian War*, the 5th century BC war between Athens and Sparta, we find the first description of chemical warfare – the formation of toxic sulfur oxide by burning sulfur. In the year 184 BC, Hannibal of Carthage used baskets with poisonous snakes against his enemy. Both Socrates and Hamlet's father were poisoned with koniin. Aqua Toffana containing arsenic was also a known poison in ancient Italy. Leonardo da Vinci proposed a powder of arsenic sulfide in the 15th century. There are many more examples of the use of chemical warfare agents (Bajgar *et al.*, 2007b). Modern history shows us that terrorists have used other chemicals, such as ricin (Bulgarian G. Markov was poisoned in 1978) or dioxin (the President of Ukraine Viktor Andriyovych Yushchenko was poisoned in 2004).

In a region of Bohemia, a ''form'' of CW was used as early as 600 years ago. It was in the year 1422 when the castle of Karlstein, the property of King Charles IV, was beseiged and 1,822 kegs containing the cesspools of the streets of Prague were hurled into the castle. Allegedly, the stench in the castle was unbearable. According to historical sources, the castle defenders were probably intoxicated with hydrogen sulfide released from the contents of the cesspools, therefore showing typical symptoms of poisoning (Bajgar, 2006).

Necessary condition: existence of toxic agent
production, processing, stockpiling, transport
(both for intentional and unintentional use)
↓
USE, RELEASE
↓ ↓
intentional/unintentional
military or local conflicts, unrestrained catastrophes, incidental events
terrorism or sabotage, failure of human factor or techniques

FIGURE 3.1. Possible reasons for a release/use of toxic chemicals.

There were some attempts to prohibit CWs by international agreement or law. Most of the early attempts were bilateral or unilateral agreements directed at the use of poisons. These included the 1675 agreement between France and Germany, signed in Strasbourg, to ban the use of poison bullets.

The first international attempt to control chemical and biological weapons took place in Brussels in 1874, when the International Declaration was signed and included a prohibition against poison and poisoned arms. In spite of the Brussels and Hague Conventions – first and second – (1899 and 1907 – signatories agreed not to use projectiles that could spread asphyxiating or deleterious gases), the world witnessed the application of chemicals in warfare to an unprecedented extent during World War I (WWI). A brief summarization of the events connected with the use/release of toxic chemicals is given in Table 3.1.

III. MILITARY USE OF CWs

The intentional use of CWs for military purposes can be found in both global and local conflicts. A typical example is the warning ''Gas! Gas!'' This was common in WWI and it is well known from the E.M. Remarque novel *All Quiet on the Western Front* where Remarque suggestively describes a chemical attack with chlorine.

During WWI, many chemicals were used including mustard, asphyxiating and irritant agents. About 45 types (27 more or less irritating and 18 lethal) of toxic chemicals were used. During the latter part of 1914, irritants were used by Germany and France; the effect was insubstantial. In late 1914, Nobel prize winner Fritz Haber of the Kaiser Wilhelm Physical Institute in Berlin (chemical synthesis of ammonium in 1918) came up with the idea of creating chlorine, although this idea of using toxic chemicals in war was expressed by Admiral Dundonald as early as 1855. Chemical warfare really began in 1915, when German troops launched the first large-scale poison gas attack at Ypres, Belgium, on April 22, using 6,000 cylinders to release 168 tons of chlorine gas, killing 5,000 British, French, and Canadian soldiers. The date is recognized as ''the birthday of modern chemical warfare'' and thereafter the belligerent parties frequently used chemical gases against each other. Phosgene was introduced by Germany late in 1915. Shortly after the first chlorine attack, the Allies had primitive emergency protective masks. In May 1916, the Germans started using diphosgene, while the French tried hydrogen cyanide 2 months later and cynogen chloride the same year. The first time mustard gas was used by German troops was July 12, 1917, and after its use near Ypres it was also called yperite.

By the end of the WWI, some 124,200 tons of chemical warfare agents (chlorine, phosgene, mustard, etc.) had been released, causing at least 1.3 million casualties of which more than 90,000 were fatal. The threat of the use of CWAs led to the development of protective means not only for humans, but also for horses and dogs. The effectivity of CWs in comparison with classic munition was evident: 1 ton of classic explosives caused 4.9 casualties; 1 ton of chemical munition caused 11.5 casualties; and 1 ton of yperite caused 36.4 casualties (Bajgar, 2006).

IV. THE PERIOD BETWEEN WWI AND WWII

The terrible casualties of CWs used during WWI, and the dangerous consequences on humans and the environment, led to the signing in June 17, 1925 of the ''Geneva Protocol for the Prohibition of the Use in War of Asphyxiating, Poisonous and other Gases and Bacteriological Methods of Warfare''. This is recognized as one of the unique and famous international treaties on the prohibition of CWs. However, it neither comprises provisions for effective verification nor prohibits development, stockpiling, and transfer of CWs. Moreover, no definition of CWs was included. Despite the provisions of the Geneva Protocol, in 1935–1936 Italian troops employed CWs during their invasion of Abyssinia (Ethiopia). This first major use of CWs after WWI came after October 3, 1935, when Mussolini launched an invasion of this country. Despite the Geneva Protocol (Italy had ratified in 1928) the Italians used mustard gas with horrible effects. Later, CWs were used between Japan and China in 1937–1945. The Japanese attacked Chinese troops with mustard gas and lewisite. The Japanese, in adition to their biological program, had an extensive CWs program and were producing agent and munitions in large quantities by the late 1930s.

V. WWII

Despite the storing and stockpiling of CWs by the great powers engaged in WWII, these fatal weapons were not practically used (except small examples) during WWII (probably because of the fear of massive retaliatory use of CWs). An example of intentional use but not in military conflict was the killing of prisoners in concentration camps in Nazi Germany. The agent first used in the camps was

TABLE 3.1. Some milestones related to the use/release of CWs and toxic chemicals

Year(s)	Event
2000 BC	Toxic smoke in China inducing sleep
4th century BC	Spartacus – toxic smoke
184 BC	Hannibal – baskets with poison snakes
1168	Fustat (Cairo) – use of ''Greek fire''
1422	Bohemia region – cesspools (H_2S)
1456	Beograde – rats with arsenic
19th century	Admiral Dundonald – proposed the use of chemicals in war
1914–1918	WWI – start of chemical war
1918–1939	Development of new CWs and protective means
June 17, 1925	Geneva Protocol
December 23, 1936	Lange and Kruger – synthesis of tabun
1940–1945	Concentration camps – cyanide
1943	Synthesis of sarin
1943	Hoffmann and Stoll – synthesis of LSD-25
1945	Kuhn – synthesis of soman
1950	V agents are begun
1961–1968	Production of VX
1961–1971	Vietnam War – herbicides (impurity dioxin)
1962	BZ was introduced into military arsenals
1970	Bicyclic phosphates considered as potential CWAs
1976	Seveso – release of dioxin
1980	Some rumors on intermediate volatility agent
1984	Bhopal incident – release of methyl isocyanate
1985	Decision on production of binary CWs
1986, 1987	Demonstration of USA CWs (Tooele) and Soviet Union CWs (Shikhany) to the CD in Geneva
1987	Production of binary CWs
1988	Halabja – use of mustard
1980–1990	Rumors of new nerve agent Novichok
1989	Conference on chemical disarmament, Paris
1991	Persian Gulf War – veteran's syndrome
1992	BZ military stocks of the USA were destroyed
1992	Finalization of the rolling text of the CWC at the CD – Geneva
1993	Signing CWC in Paris
1993	Preparatory Commission on OPCW
1994	CWs of Iraq were destroyed
1994	Aum Shinrikyo – sarin attack in Matsumoto
1995	Aum Shinrikyo – sarin attack in Tokyo
April 29, 1997	CWC – entry into force; establishment of OPCW in The Hague
2000	Research on nonlethal weapons intensified
2002	Moscow theater – Fentanyl derivatives used against terrorists
April 29, 2012	CWs of the State Parties to the CWC will be destroyed but will be prolonged

carbon monoxide, followed by the more ''effective'' hydrogen cyanide released from Zyklon B. Some experiments with aconitine-impregnated shells and some other toxic compounds including biological agents were tested on prisoners.

However, during WWII, an important step in the preparation of the most dangerous CWA was observed in Germany. In Schrader's group, organophosphates (OPs) were synthesized, primarily with the aim of obtaining more effective insecticides. Between 1934 and 1944, Schrader's team synthesized approximately 2,000 OPs including two well-known OP compounds, parathion and paraoxon. As early as 1935, the government of Nazi Germany insisted that Schrader switch the primary aim from OP insecticides to CWAs. At present, OPs are widely used in agriculture, medicine (human and veterinary), and industry. These compounds also include nerve agents (the most toxic compounds of the OP group). Nerve agents such as sarin, tabun, soman, and VX are the main compounds of CWAs. The Germans were also the greatest producers of nitrogen mustard and produced about 2,000 tons of HN-3.

Tabun was synthesized in 1936, followed by others (sarin, at the end of WWII, followed by soman) and production of these agents for the military in large quantities and their stockpiling were recognized after WWII in Dyhernfurth, Poland (e.g. stocks of tabun and some quantities of sarin). The technology was subsequently transferred to Russia and research and development of new OP nerve agents was continued. During this period British and American scientists were evaluating the toxic properties of DFP.

VI. THE PERIOD AFTER WWII AND THE COLD WAR

At the end of WWII, many Allied nations seized the chemical weapons. Most of the CW manufacturing plants in Germany were taken over and moved to Russia to new sites, e.g. the military area of Shikhany. This ''takeover'' prompted other states to begin even more research on CWs. Despite the Allies' own research into CWs, very important technologies and ''know how'' were obtained from Nazi Germany for both the USA and the former Soviet Union.

The interest in CW technology was probably one reason for the change of the future border: according to Churchill's history of WWII the proposed future boundary between Poland and Germany had been primarily agreed to consist in part of the Oder river flowing to the Baltic Sea, and its tributary, the Neisse river. Before their confluence, the Neisse consists of two branches, the East Neisse and the West Neisse. The East Neisse should be the boundary, resulting in slightly more territory for Germany. Stalin held for the West Neisse and progress was delayed. No one knows why Stalin was so insistent in this matter. The reason was probably very simple: the small town of Dyhernfurth (now Brzeg Dolny), a few kilometers north of Breslau (Wroclaw) in the disputed territory and a factory for the production of nerve agents. It was estimated that when Dyhernfurth was captured it contained stockpiles of 12,000 tons of tabun, 600 tons of sarin, and an unknown amount of soman. Presumably, the factory was dismantled, and along with their stockpiles, transported to the Soviet Union (Koelle, 1981). It has been documented that the Soviets were ready to conduct a chemical attack and their research and development of CWs was very intensified.

In the USA, during the 1950s, the chemical corporations concentrated on the weaponization of sarin. At the same time, they became interested in developing CWs that incapacitated rather than killed the targets. Mescaline and its derivatives were studied but without practical output. Five years later, a new project ''Psychochemical Agents'' (later K-agents) was established. The objective was to develop a nonlethal but potent incapacitant. Nonmilitary drugs like LSD-25 and tetrahydrocannabinol were also examined. None of these agents were found to be of military importance. The first and only incapacitant was BZ, developed in 1962; however, its stocks were destroyed in 1992 as declared by the US delegation to the Conference on Disarmament in Geneva (Document of CD, 1991). These agents, intended not to kill but to induce incapacitance, are covered under the class of nonlethal weapons (Hess *et al.*, 2005).

In the former Soviet Union, as a whole in 1940–1945 approximately 110,000 tons of first generation toxic chemicals were produced and most of them were yperite and lewisite, and irritating agents. Second generation CWs were composed of nerve agents such as sarin, soman, V agents, and to a lesser degree tabun. The development of new CWs of the third generation comprised traditional and nontraditional CWs, e.g. blister and irritant agents, and nerve gases including new types, e.g. Novichok 5, whose exact chemical structure is unknown though some assessments have been made (Bajgar, 2006); it could be a nerve agent having high toxicity. Its effects are difficult to treat using common antidotes.

An example of the unintentional use of CWs has also been observed. In March 1968, thousands of dead sheep were discovered in the Skull Valley area, Arizona, USA. This area was adjacent to the US Army's Dugway open-air testing site for CWs. Nerve gas had drifted out of the test area during aerial spraying and killed the sheep. One year later, on July 8, 1969, the Army announced that 23 US soldiers and one civilian had been exposed to sarin in Okinawa during the clearing of sarin-filled bombs (Sidell and Franz, 1997).

There are a number of examples of localized conflicts where CWs have been intentionally used but cannot be verified: e.g. in 1951–1952 in the Korean War; in 1963 the Egyptians used mustard bombs against Yemeni royalists in the Arabian peninsula; in the Indo-China War (see Vietnam War); in 1970, in Angola antiplant agents were almost certainly used; and in former Yugoslavia, there were rumors of the use of psychotomimetic agents.

A. Iraq–Iran and Afghanistan War

On September 22, 1980, Iraq launched its invasion against Iran. There has been mention of the large-scale use of CWAs in the Iran–Iraq war. In November 1983, Iran informed the United Nations that Iraq was using CWs against Iranian troops. Soon after, the use of CWs was unleashed. In addition, mustard and tabun were used. It is well known that the Iraqi Government used these agents against its own citizens, more conspicuously at Halbja in March 1988. The CWs attack was the largest against a civilian population in modern times. More than 100,000 Iranians were poisoned with CWAs; sulfur mustard was the most frequently used and has induced a number of delayed complications in Iranian veterans (pulmonary, dermal, ocular, immune system depression, reproduction, malignancy, etc.) (Afshari and Balali-Mood, 2006). Other localized conflicts involving alleged use of CWs are described in detail in an extensive review (Robinson, 1971).

The Soviet Union probably used mustard (and nerve gas) in Afghanistan. The Afghanistan war was considered the Soviet Union's ''Vietnam''. The use of CWs was described by Sidell and Franz (1997). The use of CWs by Soviet forces was also significant and has been confirmed against unprotected subjects. Despite the use of CWs, the withdrawal of Soviet troops from Afghanistan was realized at the beginning of 1989.

B. Vietnam War

After WWII, the main employment of CWs is recorded in 1961–1972 when the US Army used defoliants. The herbicide Agent Orange was used during the Vietnam War and led to the injury of more than one million Vietnamese and Americans. Agent Orange (a mixture of 2,4-dichlorophenoxy acetic acid and 2,4,5-trichlorophenoxy acetic acid) contained the chemical contaminant dioxin as an impurity which caused many deaths on both sides. There were other herbicide mixtures such as Agent White (2,4-D and picloram) and Agent Blue (cacodylic acid). The biological effects of dioxin were described by Sofronov *et al.* (2001). The first major operation of this type was conducted over the Ca Mau peninsula in September–October 1962. The area sprayed with defoliants during 1965 had been five times larger than in 1965 and in 1967 ten times larger. The scale of the use of defoliants was roughly in proportion to the overall involvement of US troops. In 1970, herbicides and defoliants were used in tens of tons, especially 2,4,5-T. The area sprayed enlarged from 23 km^2 in 1962 to 22,336 km^2 in 1969. The area exposed to spraying was assessed to be 58,000 km^2 and the number of people exposed was assessed to be more than one million including more than 1,000 deaths. In addition to defoliants used to destroy vegetation concealing the North Vietnamese, the USA used tear gas for clearing tunnels and bunkers. The irritants CS, CN, and DM were reported to be used. The total CS procured was approximately 7,000 tons from 1963 to 1969.

C. Development of VX Agent

VX was synthesized in the 1960s on the basis of the results of Tammelin and Aquilonius (Aquilonius *et al.*, 1964; Tammelin, 1957). The manufacturing of VX began in the USA in 1961. Construction of the USA's VX agent production plant at Newport, Indiana, was completed in 1961, when the first agent was produced. The production facility only operated for 7 years, and was placed on standby in 1968 (Smart, 1997).

During the same period, Soviet scientists developed the so-called Russian VX (VR, RVX, R 033) (Kassa *et al.*, 2006; Kuca *et al.*, 2006). The chemical structure of VX was unknown for a long time. Therefore some attempts to resolve this question have been made (Bajgar, 1968). Because of these ambiguities and difficulties in synthesis, model V agent [EDMM, *O*-ethyl *S*-(2-dimethylaminoethyl) methylphosphonothioate] was initially used in the Eastern Block to study antidotal treatment. Another structural analog of VX known as Chinese VX (CVX) was also developed and studied (Eckert *et al.*, 2006).

A very important step in the development in CWs has been the production of ''binary munition'', in which the final stage of synthesis of the agent from precursors is carried out in the munition (bomb, shell, or warhead) immediately before or during delivery to the target. In the 1950s, armed forces had begun looking at binary weapons. Until this time, CWs were unitary, i.e. the toxic agent was filled in the munition and then stored ready to be used. The binary concept – mixing or storing two less toxic chemicals and creating the nerve agent within the weapon – was safer during storage. The production of binary projectiles began on December 16, 1987 at the Pine Bluff Arsenal, Arkansas, USA.

D. Persian Gulf War

On August 2, 1990, Saddam Hussein sent Iraqi troops into Kuwait – allegedly in support of Kuwaiti revolutionaries who had overthrown the emirate. Iraq was known to have a large stockpile of CWs during its conflict with Iran and confirmed that they would use CWs.

President George Bush ordered US forces to be sent to Saudi Arabia at the request of the Saudi Government (Operation Desert Shield) – this was the buildup phase of the Persian Gulf War. As a consequence, in 1996, almost 60,000 veterans of the Persian Gulf War claimed certain medical problems related to their war activities, some caused by exposure to nerve agents (released after the bombing and desctruction of the sarin production facility). Unexplained ''Gulf War Syndrome'' with low-dose exposure to CWAs was suggested as a possible cause. Extensive research failed to find any single case of the problem.

However, some health effects, including alterations to the immune system 3 months after the exposure to low concentrations of sarin, were demonstrated (Kassa *et al.*, 2001, 2003). In the desert, during the fall and winter of 1990–1991, the threat of chemical warfare became very real to allied military personnel. It was demonstrated by the UN Commission that major Iraqi agents were mustard, tabun, sarin, and cyclosarin. Mustard agent was relatively pure but nerve agents were a complex mixture of the agent and degradation products. Over the period from June 1992 to June 1994, the Commission's Chemical Destruction Group destroyed 30 tons of tabun, 70 tons of sarin, and 600 tons of mustard, stored in bulk and in munitions.

Suddenly, it became clear to the whole world that there were countries that have CWs and biological weapons, and there were other countries that might obtain or produce them.

VII. UNINTENTIONAL USE OF TOXIC CHEMICALS

There are two main accidents connected with the release of toxic chemicals. In July 1976, in Seveso, Italy, more than 40,000 people were exposed to dioxin, a persistent and highly toxic chemical. The first signs were skin lesions appearing on children, and after some months there was evidence of chloracne. Health consequences have been observed from that time to the present. The Seveso accident was possibly the most systematically studied dioxin contamination incident. A similar contamination of one building of the Spolana company in Neratovice (a town in the former Czechoslovakia) was also observed (Bajgar *et al.*, 2007a). Another example, the Bhopal accident, is probably the greatest industrial disaster in history. On the night of December 2–3, 1984, water inadvertently entered the methylisocyanate storage tank (containing about 40 tons of this chemical). As a result, methylisocyanate was released into the sorrounding area. There was no warning. Many people who inhaled high concentrations of toxic gas were asphyxiated because of extensive lung damage. About 150,000 people were intoxicated (50,000 seriously poisoned) and more than 2,500 people died (Bajgar, 2006).

VIII. TERRORIST USE OF CW

Terrorists have expressed an interest in nerve agents and have deployed them in attacks on unprotected civilians (Rotenberg and Newmark, 2003). A Japanese religious cult, Aum Shinrikyo, independently manufactured numerous chemical and biological agents. The first such attack with sarin occurred in Matsumoto in 1994 and the Tokyo subway in 1995. Thousands of people were affected and dozens of people died (Nagao *et al.*, 1997; Ohtomi *et al.*, 1996; Okumura *et al.*, 1998; Yokoyama *et al.*, 1998). In Matsumoto (1994), 600 people were poisoned and hospitalized, and seven died (Morita *et al.*, 1995; Nakajima *et al.*, 1997; Yoshida, 1994). The attack in the Tokyo subway (1995) resulted in 5,500 people seeking hospital evaluation and 12 deaths (Bajgar, 2006). An interesting terroristic act was described by Tsuchihashi *et al.* (2005) – a fatal intoxication with VX administered percutaneously.

Nerve agents belong to the group of OPs. These compounds in the form of pesticides are commercially available, and are used in agriculture which can lead to professional, suicidal, or accidental intoxication. The mechanism of action, diagnosis, and treatment of intoxication with OP pesticides and nerve agents is a very hot topic at present. Moreover, some principles of the effects, diagnosis, and therapy are very similar for OP and highly toxic nerve agents, and therefore the principle of action and effective treatment can be applied in general for the civilian sector too.

The use of these chemicals was observed in Moscow in 2002. The Moscow theater hostage crisis was the seizure of a crowded theater on October 23, 2002 by about 40 armed Chechen militants who claimed allegiance to the separatist movement in Chechnya. They took 850 hostages and demanded the withdrawal of Russians from Chechnya and an end to the Chechnya war. The leader of the terrorists was 22-year-old Movsar Baraev. After two and half days of waiting, Russian forces used an unknown gas pumped into the ventilation system. Officially, 39 terrorists and at least 129 of the hostages (nine of them foreigners) were killed. Some estimates have put the civilian death toll at more than 200. It was thought that the security services used an aerosol of a chemical warfare agent, first assessed as BZ, but later it was specified that an aerosol anesthetic of the Fentanyl type was used (Bajgar and Fusek, 2006).

In the hospitals, the survivors were cut off from any communications with the outside and their relatives were not allowed to visit them. An incorrect list of hospitals for victims was released. The main problem was the lack of information about those dealing with the identification and characterization of the chemical used and the unavailability of known antidotes (e.g. naloxon) by medical staff treating the victims (Bajgar *et al.*, 2007a). It appeared from this event that there were compounds not explicitly enumerated in the CWC and therefore not controlled by this Convention. Fentanyl can be considered as a nonlethal weapon (a group of so-called calmatives) and these chemicals can also be used to incapacitate animals; of course, its use against humans is not excluded (Bajgar, 2006; Hess *et al.*, 2005).

IX. NEGOTIATIONS

Though the Cold War was continuing, the political situation led to increased activities in international negotiations. At the Conference on Disarmament in Geneva, some attempts to negotiate a ban of CWs was begun, first as the Ad hoc

Working Group, and later as the Ad hoc Committee on Chemical Weapons with the mandate to negotiate the text of a convention banning CWs.

The discussions in Geneva were more intensive from 1987 and, in 1992, the elaboration of the so-called rolling text of future CWCs was finished. During these negotiations, the text of future Conventions (''rolling text'') was enlarged: the final report (CD/342) of February 2, 1983 contained 23 pages; the same report of August 23, 1985 (CD/636) had 46 pages; and CD/952 of August 18, 1989 contained 134 pages. Simultaneously with the Geneva negotiations, in September 1989, the Memorandum of Understanding between the Governments of the United States and USSR regarding a bilateral verification experiment, data exchange related to prohibition of CWs otherwise known as the Wyoming, MT, started negotiations between two main possessors of CWs. These countries also contributed to the negotiations in Geneva: they demonstrated their CWs to the Conference on Disarmament in the USA in November 1986 (Tooele) and the USSR in October 1987 (Shikhany). The final document of the Convention is approximately 200 printed pages. The Convention was then agreed in New York at the UN General Assembly and signed in Paris in 1993. The CWC (Convention on the Prohibition of the Development, Production, Stockpiling and Use of Chemical Weapons and on their Destruction) entered into force on April 29, 1997, 180 days after the deposit of the 65th instrument of ratification of the Convention by Hungary. At this time, 87 countries ratified the CWC and became original States Parties to the Convention. Simultaneously, the Organization for Prohibition of Chemical Weapons (OPCW) in The Hague started its work of supervising the destruction of CW stocks and monitoring the world's chemical industry to prevent future misuse. There are many activities of the OPCW, e.g. training of the inspectors for control of destruction of CWs including their medical protection, research, and supported activities, solving problems due to practical implementation of the CWC, control of chemical and military facilities and other activities. Russia and the USA are unlikely to meet the final stockpile destruction deadline of April 29, 2012. By the middle of 2008, 183 Signing States and 194 recognizing States had adhered to the Convention (Davey, 2008). However, there are still States nonsignatories to the Convention. CWs have a long and ancient history. A lack of CW employment in WWII suggested that ''gas warfare'' had ended. However, further development and the utility of chemicals in Vietnam and in terrorist attacks have maintained a military interest in chemical weapons.

It is clear that the use (incidental or otherwise) of toxic chemicals has impacts in different spheres of human existence such as state structures and infrastructure, economics, psychic and public behavior, and the environment. Toxic chemicals are a great consumer of natural sources, both renewable and nonrenewable. They also consume raw materials and energy, and as a consequence cause pollution of the environment and lead to deficiency of raw materials throughout the world and therefore an unequal distribution of the world's natural sources. The impact on the psychology of humankind is also important, following either chemical wars (both global and local) or use of these chemicals by terrorists. The development of new technologies is equally important because they influence positively and negatively further human development. Research in this direction can not only contribute to ''improvement'' of chemicals to obtain more effective CWAs but also improve our knowledge in basic sciences (toxicology, neuropharmacology, etc.) and allow us to better understand physiological functions in general. It is appropriate to recall the history of cholinesterases and their inhibitors. The existence of cholinesterases was predicted by H.H. Dale in 1914, i.e. 14 years before acetylcholine was demonstrated as a natural constituent of animal tissues. This research approach was changed during WWII and cholinesterases acquired a special significance in the context of chemical warfare and nerve agents (Silver, 1974). Another publication in this area (Koelle, 1963) can be considered as the first to deal with anticholinesterase agents including CWAs – nerve agents. One can only hope that in the future the only physiological and pharmacological research will be performed in a nonmilitary framework, but that may not be the case.

X. CONCLUDING REMARKS AND FUTURE DIRECTION

The threat of the use (either military or terroristic) of CWAs (and other toxic chemicals) still exists. The military use of these agents is limited but their terroristic use is unlimited. The spectrum of these agents is very large and the ability to be prepared against the use of toxic chemicals is necessary.

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CHAPTER 4

The Tokyo Subway Sarin Attack: Toxicological Whole Truth

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Humankind has not yet experienced a full-scale sarin attack in a major modern city.

I. INTRODUCTION

The Tokyo subway sarin attack occurred in 1995, following the Matsumoto sarin attack, and served as a ''wake-up call'' for anti-NBC (nuclear, biological, and chemical) terrorism policy throughout the world. In the 10 years since the attack, efforts to combat NBC terrorism have focused on rapid and effective measures to respond to attacks employing nerve agents such as sarin.

II. SARIN TOXICITY AND MECHANISM OF ONSET

Sarin is an organophosphate compound. Within the context of chemical weapons, organophosphates are collectively referred to as ''nerve agents'', of which sarin, tabun, soman, and VX are examples. Organophosphates inhibit the enzyme acetylcholinesterase (AChE), which degrades acetylcholine (ACh), a neurotransmitter substance that acts locally on nerve synapses. Once the organophosphates bind to and phosphorylate AChE to inhibit its activity, ACh accumulates at nerve terminals, resulting in enhanced ACh activity at receptor sites. ACh effects can be functionally classified based on their site of action and can have muscarinic, nicotinic, and central nervous system (CNS) effects. These effects cause the major symptoms associated with an acute organophosphate toxicity. Muscarinic effects increase parasympathetic nerve activity and cause miosis, visual disturbances (accommodation disorder), increased salivary and bronchial secretions, bronchospasm, bradycardia, and increased gastrointestinal peristaltic activity (e.g. abdominal pain, nausea, vomiting, and diarrhea). Nicotinic effects, due to hyperstimulation of neuromuscular junctions, cause fasciculations, muscle weakness, and respiratory paralysis, and increased sympathetic nerve activity leads to miosis, sweating, tachycardia, and hypertension. CNS effects due to ACh, when severe, include anxiety, headaches, excitement, ataxia, somnolence, disorientation, coma, and seizures.

Well-known symptoms of sarin toxicity include ''miosis'', ''hypersecretion'', ''bradycardia'', and ''fasciculations''. However, the mechanism of organophosphate toxicity seems to involve conflicting actions. For example, mydriasis or miosis, and bradycardia or tachycardia may occur. Acute respiratory insufficiency is the most important cause of immediate death. Early symptoms include (1) tachypnea due to increased airway secretions and bronchospasm (muscarinic effect), (2) peripheral respiratory muscle paralysis (nicotinic effect), and (3) inhibition of respiratory centers (CNS effect), which all lead to severe respiratory insufficiency. If left untreated at this stage, death will result. Cardiovascular symptoms may include hypertension or hypotension. In more severe cases, hypotension and shock develop. Various arrhythmias can also occur, and caution is required when the QT interval is prolonged. In particular, if hypoxemia is present, fatal arrhythmias may occur with intravenous administration of atropine sulfate, which means that this drug should be given intramuscularly to victims of sarin poisoning in the ''field''. Common gastrointestinal symptoms include nausea, vomiting, and diarrhea.

An ''intermediate syndrome'' lasting 1–4 days after sarin exposure is said to exist. This is due to prolonged AChE inhibition and is associated with acute respiratory muscle paralysis, motor nerve paralysis, and cervical flexor and proximal muscle paralysis. Recumbent patients who have difficulty raising their head and neck require particular care. However, the intermediate syndrome has not been reported with nerve agent toxicity in animals or humans (Sidell, 1997), and some experts even doubt that an intermediate syndrome actually exists (De Bleecker, 1992). Others believe that the cause may be due to oral toxicity or inadequate treatment (intestinal decontamination, antidote administration, and respiratory management). In organophosphate-induced delayed neuropathy (OPIDN), seen two to three weeks after exposure, and characterized by distal muscle weakness without fasciculations, the pathophysiology is not well understood. OPIDN was first reported in the 1930s due to

contamination of Jamaican ''ginger jake'' by organophosphates. This incident (so-called ''ginger paralysis'') caused lower limb paralysis in about 20,000 victims. OPIDN symptoms have also recently been reported in Matsumoto and Tokyo subway sarin victims (Himuro *et al.*, 1998; Sekijima *et al.*, 1997). Inhibition of neuropathy target esterase (NTE) plays a role in OPIDN, but despite several basic research studies, the detailed pathophysiology has not yet been established, thus making OPIDN difficult to treat.

III. OVERVIEW OF THE TOKYO SUBWAY SARIN ATTACK

The attack took place during the morning rush hour, about 8:00 am, on March 20, 1995, the day before a holiday. The attack was carried out by members of Aum Shinrikyo to distract police from carrying out a raid on the cult's headquarters. The terrorist target was government buildings in Kasumigaseki in the heart of Tokyo. Most offices in Kasumigaseki open for business at 9:30 am, but the early morning rush hour was heavy because this was a Monday. Some believe that the time of 8:00 am was because some cult members had inside information about the government offices. The police, based on an undercover investigation, suspected that Aum Shinrikyo was manufacturing sarin for use in a terror attack, but few people, even within the police department, were aware of this. The police did not have personal protective equipment (PPE), which meant that they had to borrow PPE and receive training on use of the equipment from the Self-Defense Forces. Members of the Self-Defense Forces were alerted to some of Aum Shinrikyo's planned activities, but the general public, including healthcare providers and fire department personnel, knew nothing about their activities.

According to a subsequent police report, the terrorists placed sarin in five subway trains. Approximately 600 grams of sarin at a concentration of 33% was mixed with hexane and *N*, *N*-diethylaniline and placed in a nylon/polyethylene bag. Five terrorists then wrapped the bags in newspaper, punctured the bags with the tips of their umbrellas and left the bags on the subways. In this way the sarin seeped out of the bags and vaporized, but no other active means of dispersal were used, and in this sense, the Tokyo subway sarin attack was not really a ''full-scale'' attack.

Thus, the way in which we use the lessons learned from this attack will affect our ability to adequately deal with future terrorist attacks using sarin, which could be even greater and more serious with respect to the number of victims. Can we really assume that only 12 of the approximately 5,500 victims died because the Japanese medical system was particularly well prepared for such an eventuality? Probably not. It is more likely that the relatively low number of fatalities was due to the low concentration of sarin and passive means of dispersing it. From this perspective, the Matsumoto sarin attack one year previously was more aggressive than the Tokyo subway sarin attack. In a trial after the Matsumoto incident, it was revealed that a 70% concentration of sarin was actively volatilized using an electric heater and dispersed using an electric fan. Seven victims died and 660 were injured, giving a fatality rate of 1%. In other words, if the Tokyo subway sarin attack had been conducted using the same means as those employed in the Matsumoto sarin attack, the number of fatalities may have risen to 50 or 60. Fortunately, only 12 victims died, but this suggests that the Tokyo subway sarin attack was not a full-scale attack. In other words, mankind has not yet experienced a full-scale sarin attack in a major city.

Of the bags of sarin used in the attack, two bags were not punctured. These bags were returned to the police laboratory for analysis. At one of the subway stations on the

FIGURE 4.1. Scene from a sarin attack at Tsukiji station.

Chiyoda line, Kasumigaseki, two station employees collapsed and died on the platform after they cleaned and removed the as yet unidentified object using gloves. The actual number of victims varies depending on the source, but all confirm that 12 people died in the attack and it is generally believed that at least 5,500 victims suffered mild to serious injuries; fire fighting agencies estimate 5,642 victims, and the police, 3,796 victims. Official figures released by the subway company put the total number of victims at 5,654. This includes the 12 who died (ten passengers, two employees), those hospitalized (960 passengers, 39 employees), and those treated for minor injuries (4,446 passengers, 197 employees).

This incident was the first chemical terrorist attack in a large city. There were few first responders who could even have conceived of such an attack and would have been prepared to rapidly evacuate victims from the subway station premises. Many passengers who had difficulty walking rushed out of the trains and onto the subway platform and fell down, which in effect would have increased their exposure to sarin in the subway station. In addition, the site to which many of the victims were finally evacuated at ground level where they could lie down was in close proximity to an air exhaust vent from the subway below.

Cult members arranged to puncture all the bags containing sarin at 8:00 am, and the first call for an ambulance came at 8:09 am with the first report of a ''victim with seizures at Kayabacho Station''. After 8:15 am, the reports of victims started to increase. Around this time, the fire department received a report from Tsukiji Station stating that ''an explosion occurred and several people were injured''. Calls for ambulances eventually came from 19 subway stations, and after 8:30 am, victims, either by walking or being picked up by passing vehicles, began to pour into local clinics and hospitals. According to the Tokyo Fire Department, 5,493 people were treated at 267 medical institutions in Tokyo, 17 people were treated at 11 medical institutions outside Tokyo, and among the victims, 53 were seriously injured (Ieki, 1997). Another source states that a total of 6,185 people were treated at 294 medical institutions (Chigusa, 1995). The discrepancy in the number of victims reported by different agencies attests to some of the confusion at the time. St Luke's Hospital received the largest number of victims (640 on the day of attack). The reason for this was because of its close proximity to the Hibiya line, where there were many victims, and because of a report on television which stated that ''St Luke's Hospital has the antidote for treatment''.

IV. EMERGENCY TREATMENT OF SARIN TOXICITY

In victims of the Tokyo subway sarin attack, endotracheal intubation was not difficult. The Japanese medical literature describes the standard treatment for sarin toxicity as (A) maintain the airway, (B) assist breathing, and (C) support circulation. However, in the Matsumoto sarin attack, endotracheal intubation was more difficult in many victims because of airway hypersecretion and bronchospasm. This difference in symptoms is attributable to the higher 70% concentration and active means by which the sarin was dispersed at Matsumoto, as opposed to the 33% concentration and passive means of dispersal employed in Tokyo. Dr Fredrick Sidell (now deceased), an expert on chemical terrorism in the USA, advocated decontamination, drugs, airway, breathing, and circulation (DDABC) as the basic treatment for nerve agent poisoning. Even if the so-called ABCs of emergency treatment are followed, initial efforts to achieve adequate ventilation may be in vain. Efforts to achieve adequate ventilation should be made after at least initial administration of atropine to control airway secretions and bronchoconstriction (Sidell, 1997). If healthcare professionals learn from the Matsumoto attack, they can better recognize early parasympathetic nervous symptoms, including miosis, hypersecretion, and rhinorrhea as common symptoms of chemical terrorism due to nerve agents and institute appropriate treatment with antidotes. In large-scale disasters with many victims, treatment is often deferred in those with cardiopulmonary arrest (so-called ''black tag''). However, at St Luke's Hospital, one in three persons with cardiopulmonary arrest and two patients with respiratory arrest made a full recovery and were discharged. This high rate of recovery and return to the community is unlike that seen in other types of disasters. Therefore, if medical resources are available, all victims of a sarin attack should be aggressively treated, including cardiopulmonary resuscitation (CPR) when necessary.

The global standard for the treatment of sarin toxicity is the administration of: (1) atropine, (2) an oxime agent like PAM, and (3) diazepam (Medical Letter, 2002).

Recommended doses of atropine are 2 mg in patients with mild symptoms, primarily ocular, but without respiratory symptoms or seizures; 4 mg in patients with moderate symptoms, including respiratory symptoms such as dyspnea; and 6 mg in patients with severe symptoms, including seizures and respiratory arrest, the standard administration route for which should be intramuscular. As mentioned previously, intravenous administration of atropine in the setting of severe symptoms such as hypoxemia can induce ventricular fibrillation; thus, intramuscular administration is advised. Oxime agents such as PAM (pralidoxime methiodide, or 2-formyl-1-methylpyridinium iodide oxime) should also be given. The recommended dose for PAM in moderate and severe cases of inhalation, or for liquid exposure to a nerve agent, is 1 g by intravenous infusion over 20 to 30 min. Further continuous administration of 500 mg per hour may also be required in severe cases. Since the rate of aging of nerve agent–enzyme bond is correlated with time until administration of PAM, if the aging half-life of sarin is 5 h, then PAM must be administered before this time. The oxime of choice for sarin and VX is PAM, but HI-6 should be used

FIGURE 4.2. Sarin victims at St Luke's International Hospital.

for soman and obidoxime for tabun. Seizures are treated with diazepam. This three-drug combination, atropine, PAM, and diazepam, is the global recommendation for sarin toxicity and autoinjectors are available in several countries (Vale *et al.*, 2006).

After the Tokyo subway sarin attack, St Luke's Hospital, which treated 640 victims, used about 700 ampules of PAM and 2,800 ampules of atropine (Okumura *et al.*, 1998). This calculates out to 550 mg of PAM and 2.2 mg of atropine for each victim. The route of administration was intravenous in all cases with a total dose of atropine in severe cases 1.5 mg to 9 mg (Okumura *et al.*, 1996); doses which reflect the low concentration and passive means of sarin dispersal used in the Tokyo attack.

However, in Tokyo, no one was saved by administration of PAM, and conversely, no one died because they did not receive PAM. In other words, if ''living or dying'' was the endpoint, there was no clinical evidence that PAM was effective. The only reported finding was a more rapid return of plasma pseudocholinesterase levels to normal in some patients who received PAM as compared to those who did not. But in terms of long-term prognosis, this does not rule out the effectiveness of oxime therapy. Ideally, detailed studies are needed to evaluate the efficacy of PAM, including for long-term prognosis, but there was no sophisticated study designed in victims of the Tokyo subway sarin attack.

One piece of evidence supporting the efficacy of PAM in sarin toxicity has been the clinical benefit associated with PAM in toxicity due to organophosphorous agrochemicals. However, some experts now doubt whether such a benefit really exists. For example, Peter *et al.* (2006), using meta-analytic techniques, reevaluated the effects of oxime therapy in organophosphate poisoning. Not only did they find no beneficial effects, they also reported possible adverse effects. The Cochrane reviews for clinical evidence-based medicine reported no risk/benefit evidence for the use of oxime agents in organophosphate poisoning, but they did conclude that further detailed investigations are necessary (Buckley *et al.*, 2005).

Based on the experience of Iranian physicians who treated sarin toxicity during the Iran–Iraq war (Newmark, 2004), PAM was not available on the front lines and atropine alone was used for treatment. The doses of atropine used were considerably higher than those used in the Tokyo subway sarin attack, or that are generally recommended in the USA (Medical Letter, 2002). The Iranian protocol called for initial administration of 4 mg intravenously. If no atropine effects (improvement in dyspnea or decrease in airway secretions) were seen after 1 to 2 min, 5 mg was then administered intravenously over 5 min while heart rate was monitored. A rise in heart rate of 20 to 30 beats per min was regarded as an atropine effect. In severe cases, 20 mg to 200 mg was given. Regardless of dose, the key to saving lives, in their opinion, was how soon the atropine was administered.

Thus, treatment without the use of an oxime agent is possible. Of course, ideally, in countries where this is economically possible, treatment should use the three recommended drugs: (1) atropine, (2) an oxime agent like PAM, and (3) diazepam, and the use of autoinjectors for administration is also helpful. Unfortunately, terrorist attacks using sarin are also carried out in less economically developed countries and even if the drugs are available, considerations related to cost performance need to be considered. In this sense, preference should be given to the availability of atropine and diazepam. In other words, unless it is economically feasible, funds should be used to obtain atropine and diazepam, rather than oxime agents, whose cost–benefit ratio is still inconclusive.

V. ACUTE AND CHRONIC SYMPTOMS OF SARIN TOXICITY

Based on available data from 627 victims treated at St Luke's Hospital, symptoms in order of occurrence were: miosis 568 (90.5%), headache 316 (50.4%), visual darkness 236 (37.6%), eye pain 235 (37.5%), dyspnea 183 (29.2%), nausea 168 (26.8%), cough 118 (18.8%), throat pain 115 (18.3%), and blurred vision 112 (17.9%) (Okumura *et al.*, 1998). Cases were defined as severe for seizures or respiratory arrest requiring mechanical ventilation, moderate for respiratory distress or fasciculations, and mild for eye symptoms only. Of 640 cases reported by St Luke's Hospital, degree of intoxication was severe in five, moderate in 107, and mild in 528 victims with nicotinic effects observed in those with moderate or severe symptoms.

In the Tokyo subway sarin attack, decontamination was not performed on site, and first responders and healthcare workers initially did not wear personal protective equipment (PPE). As a result, of 1,364 fire department personnel, 135 (9.9%) became secondary victims. Official reports for police department personnel were not released, but the number of secondary exposure victims was probably similar. At St Luke's Hospital, 23% of the hospital staff became secondary victims (Okumura *et al.*, 1998). The percentage of secondary victims by hospital occupation was: nurse assistants (39.3%), nurses (26.5%), volunteers (25.5%), doctors (21.8%), and clerks (18.2%). Thus, increased contact with a primary victim increased the risk of becoming a secondary victim, with the percentage of secondary victims by hospital location being the chapel (45.8%), ICU (38.7%), outpatient department (32.4%), general ward (17.7%), and the emergency department (16.7%). The high rate of secondary victims in the chapel was attributed to poor ventilation and the large number of victims sheltered there. Because it was during the winter, victims entered the chapel fully clothed. When they removed their coats, and every time they moved, some of the sarin trapped inside the clothing probably escaped, causing secondary exposure. Fortunately, none of the secondary victims died. However, in the event that a higher concentration of sarin and more effective means of dispersion had been employed in the Tokyo attack, such as that used in Matsumoto, for example, then it is likely that fatalities would have been encountered among secondary victims.

Within the context of risk communication, the so-called ''worried-well'' patients who are concerned about having been exposed to the nerve agent, and those complaining of symptoms, even though actual exposure was unlikely, also flock to hospitals seeking treatment (Bloch *et al.*, 2007). Among patients treated at St Luke's Hospital on the day of the attack, 90.5% (568/627) had miosis (pupillary constriction), an objective finding due to sarin exposure. The remaining 9.5% were considered to be ''worried-well'' patients. As days passed by, the number of ''worried-well'' patients appeared to increase, but no actual data for this is available.

The reason for the small number of ''worried-well'' patients in the Tokyo subway sarin attack is unclear. Given the extensive coverage by the news media who mentioned that victims were flocking to St Luke's Hospital, persons without definitive symptoms, or those who were unsure whether they had been exposed but who did not want to add to the confusion, avoided going to St Luke's Hospital creating a kind of natural selection process. In addition, the target of the attack was the government buildings in Kasumigaseki at heart of Tokyo, which would have meant that many of the victims were well educated. This may also have contributed to the small number of ''worried-well'' patients. Conversely, unfamiliarity with sarin and toxic gases in general may also have contributed to the low number of ''worried-well'' patients. In either case, these observations should be reviewed from the perspective of risk communication.

Fortunately, only one victim from the Matsumoto and Tokyo subway sarin attacks has still not regained consciousness and remains in a vegetative state due to anoxic brain damage (Yanagisawa *et al.*, 2006). Sarin victims treated at St Luke's Hospital were regularly followed for the development of chronic symptoms. One year after the incident a survey was conducted, and 303 of 660 victims responded (Ishimatsu *et al.*, 1996). Forty-five percent of the respondents reported that they still experienced symptoms. Regarding physical symptoms, 18.5% of the victims still complained of eye problems, 11.9% of easy fatigability, and 8.6% of headaches. Regarding psychological symptoms, 12.9% complained of fear of subways, and 11.6% still had fears related to escaping the attack. In another survey after 3 years, 88% of the respondents reported several sequelae (Okumura *et al.*, 1999). Unfortunately, these surveys may lack objectivity and may suffer from bias. For example, the response rate may have been higher among victims still complaining of symptoms.

Murata *et al.* (1997) performed a controlled comparison study in victims 6 to 8 months after the attack, with evaluations of event-related and visual-evoked potentials (P300 and VEP), brainstem auditory evoked potentials, electrocardiographic R-R interval variability (CVRR), and scores on a posttraumatic stress disorder (PTSD) checklist. In the sarin victims, P300 and VEP (P100) latencies were significantly prolonged, and the CVRR was abnormal, indicating depression of cardiac parasympathetic nervous activity. The findings suggested persistent effects of sarin in the higher and visual nervous systems. In another study, Yokoyama *et al.* (1998a) reported a delayed effect on the vestibulocerebellar system induced by acute sarin poisoning. Yokoyama *et al.* (1998b) also reported a chronic effect on psychomotor performance. In addition, Miyaki *et al.* (2005) described the chronic effects associated with psychomotor and memory function up to 7 years after exposure.

As mentioned previously, two victims with OPIDN were reported (Sekijima *et al.*, 1997; Himuro *et al.*, 1998).

As part of a series of scientific studies sponsored by the Japanese Ministry of Health, Labor, and Welfare, Matsui *et al.* (2002) conducted two studies 7 years after the sarin attack. The first study was a case control study comparing victims treated at St Luke's Hospital with a control group. Statistical analysis showed significantly higher rates of chest pain, eye fatigue, presbyopia, eye discharge, nightmares, fear, anxiety, difficulty concentrating, and forgetfulness in the victim group. Moreover, in the victim group, there were even significantly higher rates of visual blurring, myopia, problems with focal convergence, abnormal eye sensations, flashbacks, fear of returning to the attack site, and not wanting to watch news about the attacks. The rate of PTSD, as evaluated by several diagnostic criteria, was also higher in the victim group. The second study was a cohort study comparing a group who required medical intervention after the attack with a group who did not. For lethargy, diarrhea, myopia, presbyopia, problems with focal convergence, eye discharge, and apathy, there were no significant differences between the groups, but for other evaluated parameters, scores were significantly higher in the nonintervention group. Comparison of PTSD incidence based on whether intervention was received showed that the nonintervention group had a significantly higher rate of masked PTSD. There was a higher incidence of eye symptoms in the victim group than in the nonvictim group, but there was no difference between the intervention group and nonintervention group. Thus, eye symptoms are probably long-term physical sequelae of sarin exposure. In some Matsumoto cases, persistent EEG changes without seizure activity have been reported up to 5 years (Yanagisawa *et al.*, 2006).

The results of these studies suggest some long-term effects of sarin toxicity and careful follow-up and observation are indicated in these victims.

VI. LABORATORY FINDINGS IN SARIN TOXICITY

According to inpatient records from St Luke's Hospital, the most common laboratory finding related to sarin toxicity was a decrease in plasma cholinesterase (ChE) levels in 74% of patients. In patients with more severe toxicity, plasma ChE levels tended to be lower, but a more accurate indication of ChE inhibition is measurement of erythrocyte ChE, as erythrocyte ChE (AChE) is considered ''true ChE'' and plasma ChE is ''pseudo ChE''. However, erythrocyte ChE is not routinely measured, whereas plasma ChE is included in many clinical chemistry panels; thus, it can be used as a simple index for ChE activity. In both the Matsumoto and Tokyo subway sarin attacks, plasma ChE served as a useful index of sarin exposure. In 92% of hospitalized patients, plasma ChE levels returned to normal on the following day. In addition, inpatient records from St Luke's Hospital showed an elevated creatine phosphokinase (CPK) and leukocytosis in 11% and 60% of patients, respectively. In severe cases in the Matsumoto attack, hyperglycemia, ketonuria, and low serum triglycerides due to toxicity of sarin on the adrenal medulla were observed (Yanagisawa *et al.*, 2006).

VII. CONCLUDING REMARKS AND FUTURE DIRECTION

This chapter has discussed sarin toxicity based on experiences of the attacks in Matsumoto and the Tokyo subway, and also the Iran–Iraq war. This section provides some conclusions drawn from the toxicological issues related to sarin.

Given the low concentration and means of dispersal, the Tokyo subway sarin attack can be referred to as a ''passive'' attack. The implication of such an assumption is therefore that mankind has not yet witnessed a ''full-scale'' sarin attack in any major city. While valuable information can certainly be gained from the Tokyo subway sarin attack, the experience obtained from the more aggressive Matsumoto sarin attack and the Iran–Iraq war should also be considered when developing initiatives directed at dealing with a potential ''full-scale'' attack in the future where the effects will be more serious.

Importantly, reliable epidemiologic data is lacking regarding the long-term effects of sarin toxicity, whether low dose exposure to sarin has any long-term effects, and specific effects on children, pregnant women, and fetuses (Sharp, 2006). The sporadic and limited epidemiologic surveys undertaken to date suggest that some long-term effects are present. Thus, well-designed international epidemiologic studies should be conducted in victims exposed to sarin in Japan, Iran, and during the Persian Gulf War.

There are several issues regarding treatment that need to be resolved. Before the Tokyo subway sarin attack in 1995, treatment of chemical weapons victims was exclusively regarded as a military issue; however, since then, the deliberate release of nerve agents against the general public has become a serious public safety issue. Treatment of chemical weapon injuries in a military setting assumes that one is dealing with healthy males, who have received basic and ongoing training, and who are wearing PPE. In an attack on the general public, however, we are dealing with a heterogeneous population from different backgrounds, and the victims will include women, pregnant women, children, and persons who are elderly, sick, and disabled. Furthermore, the public is defenseless against chemical weapons because of their lack of knowledge of dangerous chemical substances, or lack of experience with wearing PPE. Taken together, there is thus the potential to have thousands of victims in the event that there is a deliberate release of nerve agents against ordinary citizens.

Therefore, the medical treatment required for responding to a chemical terrorist attack on the general public will require a different strategy than that employed for such attacks in a military setting. This is because, even though there are numerous lessons that can be learned from military experience, there will be measures that may not be applicable to an attack on the public. An important issue is the means by which appropriate drugs can be safely and reliably supplied to a large number of victims. In addition, it is unrealistic to expect that first responders wearing PPE will be able to establish intravenous lines in large numbers of victims at the scene of a terrorist attack and the use of autoinjectors for intramuscular or intraosseous access is more realistic (Ben-Abraham *et al.*, 2003). In this regard, what is needed are not standardized autoinjectors issued to military personnel, but rather, a variety of autoinjectors that are uncomplicated and easy to use by victims. Research on the drugs used to treat chemical terrorism victims crosses the military/private sphere and is being conducted in several countries. However, unlike drugs that are designed for treating diseases, clinical trials cannot be performed in humans due to ethical concerns. Conducting a randomized control study is also difficult because of an insufficient number of cases of organophosphate poisoning to establish a reliable sarin toxicity model. A prime example is the oxime agent HI-6. Despite being developed more than 10 years previously, considerable time elapsed before its widespread use. From the standpoint of international security, collaborative research on drugs for treating chemical terrorism and a global agreement on standard treatment are needed. These are important issues in clinical toxicology that require international collaboration.

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CHAPTER 5

Epidemiology of Chemical Warfare Agents

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I. INTRODUCTION

While chemical warfare agents have been used for decades in military conflict, it is only in the last two decades that increasing attention has been placed on the acute and chronic health effects associated with exposure to these agents. The Gulf War of 1991 and the subsequent reports of ill-defined illnesses in the veterans of that conflict, followed by the 1995 sarin terrorist event in the Japanese subway system, placed increased attention on the capacity of deliberate or accidental exposure to chemical warfare agents resulting in significant human death and subsequent disability.

Epidemiological studies of chemical warfare agents have suffered problems in determining exposure. Other than epidemiological investigations following the Japanese terrorist event, little objective epidemiological evidence is available. In this chapter, the major studies that have been conducted on populations exposed to the chemical warfare agents are discussed and methodological issues summarized.

II. PRE-WORLD WAR II

The first full-scale deployment of chemical warfare agents was during World War I in 1915, when the Germans used chlorine gas against French, Canadian, and Algerian troops. Deaths were light, though casualties relatively heavy. A total of 50,965 tons of pulmonary, lachrymatory, and vesicant agents were deployed by both sides of the conflict, including chlorine, phosgene, and mustard gas. Official figures declare about 1,176,500 nonfatal casualties and 85,000 fatalities directly caused by chemical warfare agents during the course of the war (Heller, 2005). In 1925, 16 of the world's major nations signed the Geneva Protocol, pledging never to use gas in warfare again; however, there were subsequent reports of its use. In 1935 Italy used mustard gas during the invasion of Ethiopia in the Second Italo-Abyssinian War with 15,000 chemical casualties reported. In this military conflict and subsequent wars in which chemical agents were used, no systematic attempt was made to accurately describe the epidemiology of the exposures, nor were any accurate data established to follow the health of exposed populations after the acute exposure.

Concern regarding potential long-term effects of these exposures continued to be an issue and in 1975 a longitudinal follow-up study of the mortality experience of three samples of World War I veterans was conducted to determine if a single exposure to mustard gas with respiratory injury was associated with increased risk of lung cancer in later life (Norman, 1975). Rosters of men born between 1889 and 1893 [2,718 exposed to mustard gas, 1,855 hospitalized with pneumonia in 1918, and 2,578 with wounds of the extremities (controls)] were traced via the Veterans Administration's death records. The 4,136 deaths reported were 95% of that expected. Observed deaths from lung cancer numbered 69, or 2.5%, for the mustard-gas group as compared to 33, or 1.8%, for the pneumonia group and 50, or 1.9%, for the controls. The risk of death from lung cancer among men gassed relative to that for the controls was estimated as 1.3, with 95% confidence limits of 0.9–1.9.

III. WORLD WAR II

In 1938, the chemical structure of sarin nerve gas was discovered by the Germans, followed by the discovery of the nerve agent soman in the spring of 1944 (Schmaltz, 2006), but chemical warfare was not extensively used by either side due in part to fear of a devastating Allied retaliatory attack. There was one account of an exposure to mustard gas among Allied troops when several American ships were sunk by the Germans in 1943, including one carrying mustard gas intended for use in retaliation by the Allies if German forces initiated gas warfare. Because the presence of the gas was highly classified, authorities ashore, treating casualties, had no idea that they were seeing the effects of mustard gas and prescribed improper treatment. This incident was not uncovered for many years and military records account that 69 deaths were attributed in whole or part to mustard gas, out of 628 mustard gas military casualties (US Naval Historical Center, 1943). The due impact of the gas exposure to military and civilian populations was not accurately reported due to the high secrecy regarding the exposure and the difficulty discerning the effect of gas exposure from other types of injuries.

During the Holocaust, the Nazis used the insecticide Zyklon B containing hydrogen cyanide to kill several

million people in extermination camps and reportedly used poison gases during the Warsaw Ghetto Uprising in 1943. Human experiments were conducted on concentration camp prisoners using mustard gas and phosgene.

In 1994 a United States Senate Report, entitled ''Is military research hazardous to veterans health? Lessons spanning a half century'' reported that US military personnel were used as human subjects in the 1940s to test the chemical agents mustard gas and lewsite. This testing was done to determine how to best protect military troops from the effects of chemical warfare agents (Pechura and Rall, 1993).

During the war, the US military conducted a secret research program aimed at determining how best to protect military personnel against the effects of mustard gas and a similar compound, lewisite (Pechura and Rall, 1993). Up to 4,000 men took part in the program which required participants to wear gas masks and clothing that had been treated in an attempt to block the gas from reaching the skin. Men were required to remain in the sealed test room from 1 to 4 h. Some men were tested in the field where they were required to stay in an area that had been bombed with mustard gas anywhere from 1 h to 3 days. In 1992, the US Department of Veterans Affairs (VA) began to allow compensation for seven conditions that can result from mustard gas exposure: laryngitis, chronic bronchitis, emphysema, asthma, chronic conjunctivitis, chronic keratitis, and corneal opacities. Following publication of a report by the National Academy of Sciences (Pechura and Rall, 1993), the VA extended the list to include respiratory cancers (nasopharyngeal, laryngeal, and lung except for mesothelioma), skin cancer, chronic obstructive pulmonary disease, and acute nonlymphocytic leukemia.

In 2000, Bullman and Kang (2000) reported a 50-year mortality follow-up study of veterans exposed to low levels of mustard gas. They conducted a retrospective mortality follow-up study of World War II Navy veterans who received low-level nonlethal exposures to mustard gas while participating in mustard gas chamber tests at Bainbridge, Maryland, between 1944 and 1945. These veterans were exposed to mustard gas while wearing protective clothing and masks. Control veterans consisted of 2,663 Navy veterans who served at the same location and time as the exposed, but did not participate in chamber tests. The investigators found no excess of any cause-specific mortality associated with varying levels of mustard gas exposures that were sufficient to cause skin reactions. A significant strength of this study was that the length of time in the exposure chamber, the dose of exposure, and documentation of any observative acute effect were available for each of the exposed subjects so that a dose–response analysis could be done.

In a 2000 report, Schnurr *et al.* (2000) reported on the prevalence of current post-traumatic stress disorder (PTSD) associated with participation in these secret military tests of mustard gas exposure. Using the registry established by the VA, 363 male military veterans were randomly sampled and found to have a current prevalence of 32% for full PTSD and 10% for partial PTSD. Prevalence of PTSD varied as a function of risk and protective factors, including volunteering, physical symptoms during the tests, and prohibited disclosure. Veterans with full PTSD reported poorer physical health, a higher likelihood of several chronic illnesses and health-related disability, greater functional impairment, and higher likelihood of healthcare use than those with no PTSD. Veterans with partial PTSD also had poorer outcomes than did veterans with no PTSD in a subset of these domains.

Schnurr *et al.* (1996) postulate that these exposures involved elements of ''contamination stressors'' in which information about the exposure is the stressor rather than the tangible event. The late disclosure of the dangerous nature of these tests served as an additional stressor for many of the exposed men. Lack of information during the test, leaning to vague or diffuse fear with unknown consequences, could also contribute to the development of PTSD. The contamination stressor led to a future orientation; a worry about what problems will develop as a result of the previous exposure.

IV. POST-WORLD WAR II

Development of other agents such as the VX nerve agent continued during the 1950s and in 1961 the USA was producing large amounts of VX and performing its own nerve agent research. In 1952 the US Army patented a process for developing the powerful toxin ricin.

In 1969, 23 US servicemen and one US civilian stationed in Okinawa, Japan, were exposed to low levels of the nerve agent sarin while repainting the depot's buildings. When the exposure was publicized, the USA moved the weapons in 1971 to Johnston Atoll. Between 1951 and 1969 at the Dugway Proving Ground, various chemical and biological agents were tested. From 1962 to 1973 more than 5,800 military personnel participated in a series of tests on the vulnerability of warships to biological and chemical attacks. Only some of the involved military personnel consented to the tests. Many of the tests used chemical warfare simulants, thought at the time to be harmless. The results of the tests were reported in classified documents (SHAD report). In 2000, the Department of Defense released the names of the participants and information about the testing that occurred. In 2002 the Institute of Medicine agreed to undertake a scientific study of potential long-term health effects associated with these exposures. The IOM assembled a comparable control group and conducted a telephone health survey. Mortality records were also examined. The primary outcomes of interest were mortality, general health, and medical conditions. The SHAD participants were divided into four groups:

- Group A consisted of 3,000 participants whose exposure was limited to either *Bacillus globigii* (BG) or methylacetoacetate (MAA);
- Group B consisted of 850 participants whose only potential exposure was to trioctyl phosphate (TEHP or TOF) and contained a large number of marine participants;
- Group C consisted of 720 participants who were in tests where active chemical warfare agents were used;
- Group D consisted of 850 subjects potentially exposed to simulants who were not in groups A, B, or C.

Control groups were assembled for each of the exposed groups. Of the nearly 12,500 Navy and Marine subjects, 9,600 were assumed alive and were surveyed. The response rate for the SHAD participants was 60.8% and 46.6% for controls. No differences were observed in all-cause mortality between SHAD participants and controls, although the SHAD participants had a statistically significant higher risk of death due to heart disease. Lack of cardiovascular risk factor data makes this difference difficult to interpret. SHAD participants also reported statistically significantly worse health than controls, but no specific patterns of illness were found. Group C, the only group with potential exposure to active chemical or biological agents, reported the smallest differences in overall health compared to controls. Small differences in memory and attention as well as somatization were observed and SHAD participants had higher levels of neurogenerative conditions. SHAD participants also reported higher rates of symptoms, thought to be related to reporting bias. There were no significant differences in self-reported hospitalizations.

This report was significant in that it was the first epidemiological investigation of a military population with documented exposure to chemical agents or stimulants. The survey was conducted, however, 30 years after the exposure and with the exception of mortality records was limited to self-reported measures of health.

V. IRAN–IRAQ WAR

Saddam Hussein received chemical weapons from many countries, including the USA, West Germany, the Netherlands, the UK, France and China (Lafayette, 2002). In 1980 Iraq attacked Iran and employed mustard gas and tabun with 5% of all Iranian casualties directly attributable to the use of these agents. Iran sustained approximately 387 chemical attacks during the eight-year war (Shemirani *et al.*, 1993). About 100,000 Iranian solders were chemical warfare victims along with significant numbers of civilians. Nerve gas agents killed about 20,000 Iranian soldiers immediately. Shortly after the war ended in 1988, the Iraqi Kurdish village of Halabia was exposed to multiple chemical agents resulting in the death of 10% of the town's 50,000 residents. Hashemian *et al.* (2006) reported on the results of a cross-sectional randomized survey of 153 civilians in three towns exposed to military conflict in northwestern Iran; Oshnaviveh (low-intensity conventional warfare), Rabat (high-intensity conventional warfare), and Sardasht (both high-intensity conventional warfare and chemical weapons). The surveys measured full or partial PTSD diagnosis, anxiety symptoms, and depressive symptoms. The authors reported a 93% response rate from respondents (mean age of 45 years) and all were of Kurdish ethnicity. Compared with individuals exposed to low-intensity warfare, those exposed to high-intensity warfare and chemical weapons were at a higher risk for lifetime PTSD [odds ratio (OR), 18.6; 95% confidence interval (CI), 5.8–59.4], current PTSD (OR, 27.4; 95% CI, 3.4–218.2), increased anxiety symptoms (OR, 14.6; 95% CI, 6.0–35.6), and increased depressive symptoms (OR, 7.2; 95% CI, 3.3–15.9). Exposure to high-intensity warfare but not to chemical weapons was also significantly associated with lifetime PTSD (OR, 5.4; 95% CI, 1.7–17.6), compared with those in the low-intensity warfare group. Further, compared with individuals exposed to high-intensity warfare alone, those exposed to both high-intensity warfare and chemical weapons were at higher risk for lifetime PTSD (OR, 3.4; 95% CI, 1.5–7.4), current PTSD (OR, 6.2; 95% CI, 2.0–20.1), increased anxiety symptoms (OR, 5.6; 95% CI, 2.5–12.6), and increased depressive symptoms (OR, 3.7; 95% CI, 1.8–7.2).

This study was the first epidemiological study to document the long-term negative mental health sequelae of exposure to war and chemical weapons among civilians. The authors argue that exposure to chemical weapons is an extreme traumatic event that can result in acute helplessness and anxiety, loss of perceived safety, and chronic physical disabilities. The study had a number of limitations including the reliance on self-reported data; however, self-reported chemical exposure was verified with medical records.

VI. GULF WAR 1991

Given the past use of chemical weapons of Iraq on its own citizens, there was much concern that Saddam Hussein would again employ these weapons during the conflict against coalition forces. The only known exposure to anticholinesterase chemical warfare agents during the Gulf War was the destruction of munitions containing 8.5 metric tons of sarin/cyclosarin housed in Bunker 73 at Khamisyah, Iraq, on March 4, 1991, and additional destruction of sarin/cyclosarin rockets in a pit at Khamisyah on March 10, 1991. The US Department of Defense (DOD) reported that the exposure levels were too low to activate chemical alarms or to cause symptoms at the time of the detonation; however, several studies have been conducted to assess long-term health effects associated with this exposure. The DOD conducted modeling of the air plume that resulted from the detonation and estimated the extent of troops potentially exposed to the plume.

McCauley *et al.* (1999) conducted a computer-assisted telephone survey of 2,918 Gulf War veterans from Oregon, Washington, California, North Carolina, and Georgia to evaluate the prevalence of self-reported medical diagnoses and hospitalizations among this potentially exposed population and among comparison groups of veterans deployed and nondeployed to the Southwest Asia theater of operations. Troops reported to be within 50 km of the Khamisyah site did not differ from other deployed troops on reports of any medical conditions or hospitalizations in the nine years following the Gulf War. Hospitalization rates among deployed and nondeployed troops did not differ. Deployed troops were significantly more likely to report diagnoses of high blood pressure (OR = 1.7); heart disease (OR = 2.5); slipped disk or pinched nerve (OR = 1.5); PTSD (OR = 14.9); hospitalization for depression (OR = 5.1); and periodontal disease (OR = 1.8) when compared to nondeployed troops. There was a trend for deployed veterans to report more diagnoses of any cancer (OR = 3.0).

Smith *et al.* (2003) investigated postwar morbidity for Gulf War veterans, contrasting those who may have been exposed to low levels of nerve agents at Khamisyah and those unlikely to have been exposed. Cox regression modeling was performed for hospitalizations from all causes and hospitalizations from diagnoses within 15 categories during the period March 10, 1991 through December 31, 2000, for the duration of active-duty status. Veterans possibly exposed to nerve agents released by the Khamisyah demolition were not found to be at increased risk for hospitalizations from most chronic diseases nearly 10 years after the Gulf War. Only two of 37 models suggested that personnel possibly exposed to subclinical doses of nerve agents might be at increased risk for hospitalization from circulatory diseases, specifically cardiac dysrhythmias.

In 2005, Bullman *et al.* (2005) reported the results of a mortality study of troops exposed to chemical warfare agents based on the air plume models that were developed after the detonation. The cause-specific mortality of 100,487 exposed veterans was compared with that of 224,480 unexposed US Army Gulf War veterans. The risks for most disease-related mortality were similar for exposed and unexposed veterans. However, exposed veterans had an increased risk of brain cancer deaths (relative risk = 1.94; 95% CI = 1.12, 3.34). The risk of brain cancer death was larger among those exposed 2 or more days than those exposed 1 day when both were compared separately to all unexposed veterans.

This same team of investigators also conducted a study to examine the association of exposure to the Khamisyah plume with subsequent self-reported morbidity (Page *et al.*, 2005). The study sample included 1,056 deployed Army Gulf War veterans who responded to the 1995 National Health Survey of Gulf War Era Veterans and who were resurveyed in 2000. One-half of the subjects had been notified of potential exposure to chemical warfare agents and one-half had not. Comparing notified and nonnotified subjects, there were no statistically significant differences with respect to bed days, activity limitations, clinic visits, or hospital visits. Among 71 self-reported medical conditions and symptoms, there were five statistically significant differences, four of which were for lower rates of illness among notified subjects.

Page and colleagues also published a similar study undertaken to investigate whether possible chemical warfare exposure was associated with morbidity among Army Gulf War veterans using morbidity data for 5,555 Army veterans who were deployed to the Gulf region (Page *et al.*, 2005). Responses to 86 self-assessed health measures, as reported in the 1995 Department of Veterans Affairs National Health Survey of Gulf War Era Veterans, were evaluated. They found little association between potential exposure and health, after adjustment for demographic variables. The investigators concluded that potential exposure to sarin or cyclosarin at Khamisyah did not seem to have adversely affected self-perceived health status, as evidenced by a wide range of health measures.

More recently, Heaton examined the association between modeled estimates of sarin/cyclosarin exposure levels and volumetric measurements of gross neuroanatomical structures in 1991 Gulf War veterans with varying degrees of possible low-level sarin/cyclosarin exposure (Heaton *et al.*, 2007). Twenty-six GW-deployed veterans recruited from the Devens Cohort Study participated. Magnetic resonance images of the brain were acquired and analyzed using morphometric techniques, producing volumetric measurements of white matter, gray matter, right and left lateral ventricles, and cerebrospinal fluid. Volumetric data were analyzed using exposure estimates obtained from refined models of the 1991 Khamisyah presumed exposure hazard area. No differences were observed in the 13 ''exposed'' veterans when compared to 13 ''non-exposed'' veterans in volumetric measurements of discrete brain tissues. However, linear trend analyses showed a significant association between higher levels of estimated sarin/cyclosarin exposure and both reduced white matter (adjusted parameter estimate = 4.64, $p < 0.0001$) and increased right lateral ventricle (adjusted parameter estimate = 0.11, $p = 0.0288$) and left lateral ventricle (adjusted parameter estimate = 0.13, $p < 0.0001$) volumes. These findings suggest subtle but persistent central nervous system pathology in Gulf War veterans potentially exposed to low levels of sarin/cyclosarin.

This investigative team also compared previous neurobehavioral performance results collected prior to notification of veterans who were potentially exposed in the Khamisyah detonation (Proctor *et al.*, 2006). They hypothesized the exposure to sarin and cyclosarin would be associated with poorer performances on objective neurobehavioral tasks in specific functional domains (particularly in visuospatial abilities and psychomotor functioning) in a dose-dependent manner. They found that sarin and cyclosarin exposure was significantly associated with less

proficient neurobehavioral functioning on tasks involving fine psychomotor dexterity and visuospatial abilities 4–5 years after exposure. They concluded that the findings suggest a dose–response association between low-level exposure to sarin and cyclosarin and specific functional central nervous system effects 4–5 years after exposure.

VII. TERRORISM

Two terrorist attacks with the nerve agent sarin affected populations in Matsumoto and Tokyo, Japan, in 1994 and 1995 killing 19 and injuring more than 6,000. Morita *et al.* (1995) described the acute effects including instantaneous death by respiratory arrest in four victims in Matsumoto. In Tokyo, two died in station yards and another ten victims died in hospitals within a few hours to 3 months after poisoning. Six victims with serum cholinesterase (ChE) below 20% of the lowest normal were resuscitated from cardiopulmonary arrest (CPA) or coma with generalized convulsion. Five recovered completely and one remained in a vegetative state due to anoxic brain damage. EEG abnormalities were observed for up to 5 years in certain victims. Miosis and copious secretions from the respiratory and gastrointestinal tracts (muscarinic effects) were common in severely to slightly affected victims. Weakness and twitches of muscles (nicotinic effects) appeared in severely affected victims. Neuropathy and ataxia were observed in a small number (less than 10%) of victims, in which findings disappeared between 3 days and 3 months. Leukocytosis and high serum CK levels were common. Hyperglycemia, ketonuria, low serum triglyceride, and hypopotassemia were observed in severely affected victims, in which abnormalities were attributed to damage of the adrenal medulla.

The Matsumoto Japanese government assembled a committee of city government, local hospitals and physicians from Shinsu University to monitor the immediate and long-term effects of the exposure, resulting in the most comprehensive epidemiological studies of acute and residual effects of exposure to chemical warfare agents. Three weeks after the attack, community residents ($n = 2{,}052$) residing in an area within 1,000 to 850 m of the attack were surveyed and categorized as severely affected (admitted to the hospital), moderately affected if treated in outpatient clinics, and slightly affected if they had symptoms but did not seek medical attention. At the time of this follow-up survey, 28% of the affected residents remained symptomatic (69% of the severely affected, 42% of the moderately affected, and 14% of the slightly affected). The most frequent persisting symptoms were fatigue, dysesthesia of extremities, and ocular pain. Visual problems continued in about 10% of severely affected victims (Yanagisawa *et al.*, 2006).

In the Tokyo subway attack, 640 victims were seen within hours of the incident. Five were critically injured and required mechanical ventilation. One hundred and seven were moderately injured with systemic symptoms and signs of respiratory, digestive, and/or neurological systems in addition to ocular signs. The large majority ($n = 528$) had only eye signs or symptoms and were released after several hours of observation (Yanagisawa *et al.*, 2006).

There have been a number of investigations of the health of the survivors of the Tokyo subway attack. Yokoyama *et al.* (1998) conducted a study of 18 victims 6–8 months after the attack. At that time the mean plasma ChE was 72.1, lower than the ''normal'' range of 100–250 IU/l. In neurobehavioral testing at that time, sarin cases had significantly lower scores on the digit symbol test than the control group. Scores on the General Health Questionnaire and fatigue were significantly higher in the victims and PTSD scores were also increased. Postural balance was also different in victims suggesting that integration of visual input might have been impaired. P300 and VEP (P100) latencies in the sarin cases were significantly prolonged compared with the matched controls (Murata *et al.*, 1997). In the sarin cases, the CVRR was significantly related to serum ChE levels determined immediately after exposure; the PTSD score was not significantly associated with any neurophysiological data despite the high PTSD score in the sarin cases. These findings suggest that asymptomatic sequelae to sarin exposure, rather than PTSD, persist in the higher and visual nervous systems beyond the turnover period of ChE.

The National Police Academy (1999) conducted a survey of 1,247 residents who reported to the Police Department that they had contact with sarin at the incident. More than half complained of physical symptoms, such as asthnopia and decrease in visual acuity. Seventeen percent reported psychological trauma from the event with 14% still unable to ride on subways 3 years after the incident.

There continued to be follow-up studies indicating the residual effects of the attack. Ohtani *et al.* (2004) followed 34 victims 5 years after the attack. Not only PTSD but also nonspecific mental symptoms persisted in the victims at a high rate. A total of 11 victims were diagnosed with current or lifetime PTSD. Victims with PTSD showed higher anxiety levels and more visual memory impairment.

Yamasue *et al.* (2007) conducted a 5 year follow-up study to identify persistent morphological changes subsequent to the attack. Thirty-eight victims of the sarin attack, who had been treated in the emergency department for sarin intoxication and 76 matched health control subjects underwent weighted and diffusion tensor magnet resonance imaging. ChE values were compared to levels immediately after the attack. The voxel-based morphometry exhibited smaller than normal regional brain volumes in the insular cortex and neighboring white matter, as well as in the hippocampus in the victims. The reduced regional white matter volume correlated with decreased serum cholinesterase levels and with the severity of chronic somatic complaints related to interoceptive awareness.Voxel-based

analysis of diffusion tensor magnetic resonance imaging further demonstrated an extensively lower than normal fractional anisotropy in the victims. These findings suggest that sarin intoxication might be associated with structural changes in specific regions of the human brain.

Rescue and safety workers have also been studied. Nishiwaki *et al.* (2001) studied 27 male rescue team staff and 30 police officers, 3–45 months after the event. The study subjects showed decreased performance on the digit span test; however, no effects on stabilometry and vibration perception threshold were found. Li *et al.* (2004) followed 27 male firefighters and 25 male police officers three years after the attack for genotoxic effects. They found an elevated frequency of sister chromatid exchanges in lymphocytes of the victims which were related to the percentage of ChE inhibition observed just after the attack.

VIII. CONCLUDING REMARKS AND FUTURE DIRECTION

This chapter described the major epidemiological studies of populations who have been exposed to chemical warfare agents. Many of the studies of military populations have suffered from inaccurate exposure assessment and lack of clinical data. The studies in the past decade of the survivors of the sarin terrorist attacks provide the most comprehensive data to date on the scope of health outcomes associated with these exposures. These reports point to the need for long-term follow-up studies of victims following such events. The data from the terrorist events and the Gulf War when many troops believed they were exposed to chemical agents point to the prevalence of PTSD associated with real or threatened exposure.

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