Introduction, Review of Literature and Objective of Work

1. Introduction

One of the greatest concerns in modern world is the possibility of use of chemical weapons by regular forces or by terrorist groups.\(^1\) Chemical Warfare is the use of the toxic chemical to kill, injure or incapacitate an enemy in warfare and associated military operations. A chemical substance intended for such use in military operations is defined as a Chemical Warfare Agents (CWAs). The CWAs are the most ancient and most dangerous weapons of mass destruction. These CWAs historically utilized in an effort to gain military superiority. The probability of use of CWAs as weapon of mass destruction is very significant because they can be easily synthesized by simple chemical reactions and they possess extreme toxicity. Compared to conventional weaponry, relatively small amounts of chemical agents may cause high numbers of casualties. A chemical for use as a CWAs it has to meet a number of stringent tactical and technical requirements before qualifying as a chemical warfare agent, like high toxicity, rapidity of action, persistency, penetration capability, suitable vapor pressure etc.

The modern era of potent CWAs utilization is considered to begin with the weaponization of chlorine and phosgene gases in 1915 by the German army.\(^2\) By the end of the world war I, attacks with above toxic agents had inflicted roughly one million casualties, about 90000 of them are fatal. The first Nerve agents (Tabun) were developed by German during the World War II. During Iraq-Iran war from 1980-1988, Iraq used nerve agents (sarin and tabun) and blister agent (sulphur mustard) against Iran, killing hundreds of military and civilian populations. The destructive and non-discriminating nature of CWAs has led to recent attempts by world bodies to restrict the development and destroy existing stockpiles of these agents.
2. **History of Chemical Warfare Agents**

The use of CWAs can be retraced to 1,000 B.C., when the Chinese employed smokes containing arsenic, while the Greek utilized chemical compounds to poison water and other supplies of their enemies. With the progress of time different CWAs are developed as and when required, some of them are explained as follows. Carl Scheele, a Swedish chemist, was credited with the discovery of chlorine in 1774. He also determined the properties and composition of hydrogen cyanide in 1782. Comte Claude Louis Berthollet, a French chemist, synthesized cyanogen chloride in 1802. Sir Humphry Davy, a British chemist, synthesized phosgene in 1812. Sulphur Mustard was synthesized in 1822. John Stenhouse, a Scotch chemist, synthesized chloropicrin in 1848. Important historical facts about the use of chemical weapons are presented in the following table.

**Table 1: Historical facts about the use of CWAs**

<table>
<thead>
<tr>
<th>Date</th>
<th>Historical fact</th>
</tr>
</thead>
<tbody>
<tr>
<td>1915-18</td>
<td>Use of chemical weapons in World War I.</td>
</tr>
<tr>
<td>1935</td>
<td>Italy uses mustard gas in Libya and Ethiopia.</td>
</tr>
<tr>
<td>1936</td>
<td>The German chemist Gerhard Schrader synthesized the neurotoxic agent tabun.</td>
</tr>
<tr>
<td>1937</td>
<td>Schrader and co-workers synthesized the neurotoxic agent sarin</td>
</tr>
<tr>
<td>1939</td>
<td>Japan uses mustard gas against China.</td>
</tr>
<tr>
<td>1940-45</td>
<td>Germany employed Zyklon B, a variant of HCN gas chambers</td>
</tr>
<tr>
<td>1942</td>
<td>Germany begins the industrial production of nerve gases.</td>
</tr>
<tr>
<td>1944</td>
<td>The German chemist R. Kuhn synthesized soman.</td>
</tr>
<tr>
<td>1950</td>
<td>VX is synthesized by the British &amp; weaponized by USA.</td>
</tr>
<tr>
<td>1984-86</td>
<td>The use of CWAs by Iraq in the war against Iran is conformed.</td>
</tr>
<tr>
<td>1988</td>
<td>The use of chemical weapons by Iraq in the repression against the Kurdish population</td>
</tr>
<tr>
<td>1994</td>
<td>Terrorist attack with sarin in Matsumoto, Japan, executed by the Aum Shinrikyo, results in 7 deaths and more than 300 injured</td>
</tr>
<tr>
<td>1995</td>
<td>Aum Shinrikyo uses sarin to attack the Tokyo subway, causing 12 deaths and thousands of casualties</td>
</tr>
</tbody>
</table>
Today it is considered that modern chemical war began during the First World War, more precisely on April 22nd 1915, when the German Army employed large quantities of chlorine gas against the Allied Forces in Ypres, Belgium. British and French troops equally retaliated, and the First World War became the first stage of the use in large scale of several poisonous gases, like phosgene, diphosgene, hydrogen cyanide, cyanogen chloride and mustard gas. Japan also used CWAs against the Chinese forces between the end of the 1930’s and the beginning of the 1940 during second World War.

During the Second World War, Saunders synthesised organophosphorus esters, which possess nerve-poisoning effects. The nerve agents are most lethal organophosphorus compounds of a type first synthesized in 1854, but their development as warfare agents did not occur until eight decades later. The first of military nerve agents, known as Tabun or GA was synthesised in 1936 by Schrader while he was searching for more effective agricultural organophosphate insecticides at German chemical industry. Two years later, he synthesised second nerve agent known as Sarin or GB. In 1944, a third nerve agent, now known as Soman or GD, was synthesised. Because of their unique properties and high toxicity of these nerve agents, they have attracted the attention of many nations and became a threat.\textsuperscript{7-8}

In 1950, Dr. Ranjeet Ghos and J.F. Newman discovered new organophosphorous agent Amitone (VG) at imperical chemical at United kingdom. Recently in August 2013, rebellion groups used nerve agent (Sarin) in Damascus, Syria, hundreds of people were killed.

3. Classification of Chemical Warfare Agents

The chemical warfare agents possess different characteristics and belong to various classes of compounds with pronounced physiological and chemical properties.\textsuperscript{9} Different chemical warfare agents cause different symptoms and injuries to their victims. Because of this range of potential effects, identifying the chemical agent is a key step to determining the most effective treatment. Also, chemical weapons may produce their effects by multiple different exposure routes, for example by skin contact or by inhalation. Based on these factors chemical warfare agents are classified as follows.\textsuperscript{10}
3.1 Nerve Agent

These are organophosphorous compounds which inhibit the enzyme acetylcholinesterase (AchE). Nerve agents, interfere with the central nervous system by reacting with the enzyme acetylcholinesterase and creating an excess of acetylcholine which affects the transmission of nerve impulses. The symptoms of nerve agent poisoning includes difficulty in breathing, drooling and excessive sweating, vomiting, cramps, involuntary defecation and urination, twitching, jerking and staggering, headache, confusion, drowsiness, convulsion, coma, dimness of vision and pinpointing of the pupils. Most nerve agents (Fig. 1) belong to a group of chemicals called organophosphates.

![Figure 1: Structure of important nerve agents and their analogues](image)

These agents are mainly liquid in nature. These are categorized as G series agents: GB (Sarin), GD (Soman), GA (Tabun), GF and V Series agents: VE, VG, VM and VX, the letter “G” representing the country of origin “Germany” and letter “V” possibly denoting “Venomous”. Nerve agent poisoning may be treated with timely administration of antidotes such as atropine and diazepam.
3.2 Blister Agent

Chemicals categorized as blister agents, also known as vesicants, cause painful blistering of the skin (Fig. 2). These are not as lethal as nerve agents. These chemicals cause incapacitation rather than death but can kill in large doses. These agents are very persistent in the environment. The most common blister agents are called mustard agent, as it smells like mustard. Mustard agents are oily liquids that range in color from very pale yellow to dark brown, depending on the type and purity, and have a faint odour of mustard, onion, or garlic. These liquids evaporate quickly, and their vapours are also injurious. Exposure to blister agent can cause a number of life-threatening symptoms, including severe pain, irritation and skin with large fluid blisters that heal slowly and may become infected. Mild respiratory distress is also a marked symptom on exposure to these agents. All blister agents are heavier than air, and are readily absorbed through the eyes, lungs, and skin. Effects of the two mustard agents are typically delayed: exposure to vapors becomes evident within 4 to 6 hours, and skin exposure in 2 to 48 hours.

![Figure 2: Effect of Sulfur Mustard Injury](image)

The toxicity of mustard is due to its alkylating efficiency. Most of all it binds to nucleophiles such as nitrogen in the base components of nucleic acids and sulfur in SH-groups in proteins and peptides. Mustard agent can destroy a large number of different substances in the cell by means of alkylation and thereby influence numerous processes in living tissue.
Blister agents are classified into further three groups:

(a) Sulfur mustards – A family of sulfur-based agents, including the so-called "mustard gas".

(b) Nitrogen mustards – A family of agents similar to the sulfur mustards, but based on nitrogen instead of sulfur. They are again three types as HN1, HN2, and HN3.

(c) Lewisite – These agents are similar to the above classes except that they contain arsenic in place of sulfur or nitrogen. They are again three types as L-1, L-2 and L-3.
3.3 Blood Agents
The blood agents are substances that block oxygen utilization or uptake from the blood, causing rapid damage to body tissues.\textsuperscript{14} This category includes hydrogen cyanide and cyanide salts. Hydrogen cyanide is a very volatile gas, smelling of almonds, while cyanide salts are odourless solids. Symptoms are irritation of the eyes and respiratory tract, nausea, vomiting and difficulty in breathing.

\begin{center}
\begin{tabular}{ccc}
\text{HC} & \text{N} \\
Hydrogen cyanide & & \text{Cl} & \text{N} \\
& Cyanogen chloride & \\
\end{tabular}
\end{center}

Figure 5: Different types of Blood agent

In general hydrogen cyanide is used as a chemical warfare agent. The main problem associated with this agent to be used as was gas is its quick disperse property. They are highly volatile, so that its effective concentrations are difficult to achieve on the battleground, and at the same time it is very difficult to maintain high concentration for long time in battleground. However, at high concentrations cyanide kills quickly. Potential agents are hydrocyanic acid and cyanogen chloride.

3.4 Chocking agents
The choking agents cause physical injury to the lungs through inhalation. Membranes may swell and lungs become filled with liquid, and in serious cases, due to lack of oxygen causes death. These agents are generally gases and marked odour with colour formation in the surrounding air. The examples are chloropicrin, perfluoroisobutylene (PFIB) and phosgene.

\begin{center}
\begin{tabular}{ccc}
\text{Cl} & \text{O} & \text{Cl} \\
Phosgene & & \text{Cl} & \text{C} & \text{N} & \text{O}^{-} \\
& Chloropicrin & & \text{Cl} & \text{O} & \text{Cl} \\
& PFIB & & \text{F} & \text{F} & \text{F}
\end{tabular}
\end{center}

Figure 6: Different types of chocking agent
3.5 Incapacitating agent

These are generally non-lethal agents that cause temporary physical or mental incapacitation rather than death. On exposure of these agent individual incapable of concerted effort in the performance of their assigned duties.

![3-Quinuclidinyl benzilate (BZ)](image)

Figure 7: 3-Quinuclidinyl benzilate (BZ)

3.5 Physical Properties of Chemical Warfare Agents

Some of the physical properties of different types of nerve and blister agents are presented in the following table.\(^{15}\)

Table 2: Physical properties of Nerve agent

<table>
<thead>
<tr>
<th>Sl. No</th>
<th>Parameter</th>
<th>VX</th>
<th>GA (Tabun)</th>
<th>GB (Sarin)</th>
<th>GD (Soman)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Chemical formula</td>
<td>(\text{C}<em>{11}\text{H}</em>{26}\text{NO}_2\text{PS})</td>
<td>(\text{C}_2\text{H}_8\text{NO}_2\text{P})</td>
<td>(\text{C}<em>5\text{H}</em>{11}\text{FO}_2\text{P})</td>
<td>(\text{C}<em>7\text{H}</em>{16}\text{FO}_2\text{P})</td>
</tr>
<tr>
<td>2</td>
<td>CAS No.</td>
<td>50782-69-9</td>
<td>77-81-6</td>
<td>107-44-8</td>
<td>506-77-4</td>
</tr>
<tr>
<td>3</td>
<td>Molecular weight</td>
<td>267.4</td>
<td>162.1</td>
<td>140.1</td>
<td>182.2</td>
</tr>
<tr>
<td>4</td>
<td>Physical state</td>
<td>Oily Liquid</td>
<td>Oily Liquid</td>
<td>Liquid</td>
<td>Liquid</td>
</tr>
<tr>
<td>5</td>
<td>Colour</td>
<td>Light amber</td>
<td>Colourless</td>
<td>Colourless</td>
<td>Colourless</td>
</tr>
<tr>
<td>6</td>
<td>Melting point (^{\circ}\text{C})</td>
<td>-39</td>
<td>-50</td>
<td>-56</td>
<td>-42</td>
</tr>
<tr>
<td>7</td>
<td>Boiling Point/(^{\circ}\text{C})</td>
<td>298</td>
<td>230</td>
<td>158</td>
<td>198</td>
</tr>
<tr>
<td>8</td>
<td>Density at 20(^{\circ}\text{C})</td>
<td>1.008</td>
<td>1.073</td>
<td>1.102</td>
<td>1.022</td>
</tr>
<tr>
<td>9</td>
<td>Water solubility at 25(^{\circ}\text{C}) g/L</td>
<td>30</td>
<td>98</td>
<td>Miscible</td>
<td>21</td>
</tr>
<tr>
<td>10</td>
<td>Volatility mg/m(^3)</td>
<td>10.5</td>
<td>610</td>
<td>22000</td>
<td>3900</td>
</tr>
<tr>
<td>11</td>
<td>Persistency in soil</td>
<td>2-6 day</td>
<td>1-1.5 day</td>
<td>2-24 h</td>
<td>1-3 day</td>
</tr>
</tbody>
</table>
Table 3: Physical properties of Blister agent

<table>
<thead>
<tr>
<th>Sl. No</th>
<th>Parameter</th>
<th>HD</th>
<th>HN1</th>
<th>HN2</th>
<th>HN3</th>
<th>Lewisite</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Chemical formula</td>
<td>C₄H₈Cl₂S</td>
<td>C₆H₁₃Cl₂N</td>
<td>C₅H₁₈Cl₂N</td>
<td>C₆H₁₂Cl₁N</td>
<td>C₂H₂Cl₁As</td>
</tr>
<tr>
<td>2</td>
<td>CAS No.</td>
<td>505-60-2</td>
<td>538-07-8</td>
<td>51-75-2</td>
<td>555-77-1</td>
<td>541-25-3</td>
</tr>
<tr>
<td>3</td>
<td>Molecular weight</td>
<td>159.08</td>
<td>170.08</td>
<td>156.07</td>
<td>205.54</td>
<td>207.35</td>
</tr>
<tr>
<td>4</td>
<td>Physical state</td>
<td>Oily</td>
<td>Liquid</td>
<td>Liquid</td>
<td>Liquid</td>
<td>Liquid</td>
</tr>
<tr>
<td>5</td>
<td>Colour</td>
<td>Clear/ Pale Yellow</td>
<td>Pale yellow</td>
<td>Pale yellow</td>
<td>Pale yellow</td>
<td>Colourless/) amber(Pure)/ Brown (aged</td>
</tr>
<tr>
<td>6</td>
<td>Melting point / °C</td>
<td>-13</td>
<td>-34</td>
<td>-60</td>
<td>-3.7</td>
<td>-18</td>
</tr>
<tr>
<td>7</td>
<td>Boiling Point/ °C</td>
<td>216</td>
<td>Decompose</td>
<td>75</td>
<td>240</td>
<td>190</td>
</tr>
<tr>
<td>8</td>
<td>Density at 20 °C</td>
<td>1.27</td>
<td>1.09</td>
<td>1.12</td>
<td>1.24</td>
<td>1.89</td>
</tr>
<tr>
<td>9</td>
<td>Water solubility</td>
<td>0.92</td>
<td>12</td>
<td>Sparingly soluble</td>
<td>0.16</td>
<td>0.5</td>
</tr>
<tr>
<td></td>
<td>at 25 °C g/L</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>10</td>
<td>Volatility mg/m³</td>
<td>920</td>
<td>2.29</td>
<td>3.36</td>
<td>0.12</td>
<td>4.480</td>
</tr>
</tbody>
</table>

3.6 Characteristics, Exposure Symptoms of CWAs

Different type of chemical warfare agents shows different symptoms in the body after exposed. These are presented in the flowing table.¹⁶

Table 4: Characteristics of different CWAs

<table>
<thead>
<tr>
<th>Agent classes</th>
<th>Characteristics</th>
<th>Exposure Symptoms</th>
<th>Agent Example</th>
</tr>
</thead>
<tbody>
<tr>
<td>Nerve agents</td>
<td>Attack nervous system, can enter body through inhalation or skin.</td>
<td>• Pinpoint pupils</td>
<td>Tabun, Sarin, Soman, VX</td>
</tr>
<tr>
<td></td>
<td></td>
<td>• Runny nose</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>• Tightness in chest</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>• Muscle twitching</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>• Coma</td>
<td></td>
</tr>
<tr>
<td>Blister Agent</td>
<td>Attack skin and absorbed rapidly into skin</td>
<td>• Tearing of eyes</td>
<td>Sulphur mustard, Lewisite</td>
</tr>
<tr>
<td></td>
<td></td>
<td>• Initial redness of skin, followed by blisters</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>• Pain</td>
<td></td>
</tr>
<tr>
<td>Chocking agent</td>
<td>Attack respiratory tract</td>
<td>• Irritated eyes</td>
<td>Phosgene, Chloropicrine</td>
</tr>
<tr>
<td></td>
<td></td>
<td>• Shortness of breath</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>• Pulmonary edema</td>
<td></td>
</tr>
<tr>
<td>Blood agent</td>
<td>Attack circulatory system</td>
<td>• Loss of consciousness</td>
<td>Hydrogen cyanide, Cyanogen chloride</td>
</tr>
<tr>
<td></td>
<td></td>
<td>• Headache</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>• Breathing problem</td>
<td></td>
</tr>
</tbody>
</table>
4. Toxicity of CWAs

CWAs and their hydrolysis products are extremely toxic. Organophosphorous nerve agents are showing their effect on acetylcholinesterase enzymes, where the nerve agent phosphorylate a serine hydroxyl group in the active site of the enzyme. The normal function of acetylcholinesterase is to destroy acetylcholine in the synaptic branched or neuromuscular junction, in order to terminate transmission of nerve impulse, failure of acetylcholinesterase activity results in accumulation of acetylcholine, which in turn cause enhancement and prolongation of cholinergic effects such as such as myosis, salivation, hypotension, bradycardia, muscle tremors, convulsions, and respiratory depression. A fatal outcome of the intoxication is usually due to respiratory failure. Reactivation of acetylcholinesterase occurs by dephosphorylation, and the rates of phosphorylation and dephosphorylation are very variable, which partly accounts for differences in acute toxicity between the nerve agents.\(^\text{17-18}\)

The toxicity of CWAs is measured as an estimate of the dose that can have lethal effects on humans. Lethal Dose \(_{50}\) (LD\(_{50}\)) is the estimated dose at which 50% of the population will die if exposed to CWAs. Lethal concentration \(_{50}\) (LC\(_{50}\)) and lethal concentration over time \(_{50}\) (LCt\(_{50}\)) values represents the concentration of the agent in air and inhalational toxicity of the vapor form of a volatile agent, respectively, necessary to kill 50% of the test subjects. Significance of LD\(_{50}\) is that it tells about the ‘toxic potential’ of a substance i.e. lower the LD\(_{50}\), higher is the toxicity of a compound. The toxicity data of some of CWAs are presented in the following table.\(^\text{19}\)
Table 5: Toxicity data of some common chemical warfare agents

<table>
<thead>
<tr>
<th>Agents</th>
<th>LC₅₀ₐₜ (mg min/m³)</th>
<th>LC₅₀ (mg/individual)</th>
<th>LD₅₀ (mg/Kg)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tabun (GA)</td>
<td>200</td>
<td>4000</td>
<td>1500</td>
</tr>
<tr>
<td>Sarin (GB)</td>
<td>100</td>
<td>1700</td>
<td>1700</td>
</tr>
<tr>
<td>Soman (GD)</td>
<td>50</td>
<td>300</td>
<td>350</td>
</tr>
<tr>
<td>Vx</td>
<td>6</td>
<td>8</td>
<td>10</td>
</tr>
<tr>
<td>Sulphur Mustard (HD)</td>
<td>1500</td>
<td>10000</td>
<td>100</td>
</tr>
<tr>
<td>Nitrogen Mustard (HN-1)</td>
<td>1200</td>
<td>100000</td>
<td>7000</td>
</tr>
</tbody>
</table>

A common feature of the four nerve agents is the presence of a stereogenic phosphorus atom, which leads to the presence of equal amounts of optical isomers in the synthetic product. The stereoisomers of nerve agents have different toxicological properties are presented in the following table.²⁰

Table 6: Acute Lethality of Nerve Agent Stereoisomers

<table>
<thead>
<tr>
<th>Sl. No</th>
<th>Nerve agent stereoisomer</th>
<th>LD₅₀ (mouse) µg/ kg</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>C(+)-P(-)-soman</td>
<td>99</td>
</tr>
<tr>
<td>2</td>
<td>C(-)-P(-)-soman</td>
<td>38</td>
</tr>
<tr>
<td>3</td>
<td>C(+)-P(+)-soman</td>
<td>&gt;5000</td>
</tr>
<tr>
<td>4</td>
<td>C(-)-P(+)-soman</td>
<td>&gt; 2000</td>
</tr>
<tr>
<td>5</td>
<td>C(+)-P(+)-soman</td>
<td>156</td>
</tr>
<tr>
<td>6</td>
<td>(-)-sarin</td>
<td>41</td>
</tr>
<tr>
<td>7</td>
<td>(+)-sarin</td>
<td>---</td>
</tr>
<tr>
<td>8</td>
<td>(+)-sarin</td>
<td>83</td>
</tr>
<tr>
<td>9</td>
<td>(-)-tabun</td>
<td>119</td>
</tr>
<tr>
<td>10</td>
<td>(+)+ tabun</td>
<td>837</td>
</tr>
<tr>
<td>11</td>
<td>(+)+ tabun</td>
<td>208</td>
</tr>
<tr>
<td>12</td>
<td>(-)-vx</td>
<td>165</td>
</tr>
<tr>
<td>13</td>
<td>(+)-vx</td>
<td>12.6</td>
</tr>
<tr>
<td>14</td>
<td>(+)-vx</td>
<td>20.1</td>
</tr>
</tbody>
</table>
5. **Chemical Weapons Convention (CWC)**

The Convention on the prohibition of the development, production, stockpiling and use of Chemical Weapons and their destruction, known as Chemical Weapons Convention (CWC), was opened for signature in Paris, France on 13 January 1993. The Convention had been the subject of nearly twenty years of negotiation with the aim to finalize an international treaty banning chemical weapons, and designed to ensure their worldwide elimination. The CWC entered into force on 29 April 1997, which is an international multidisciplinary treaty that prohibits the proliferation of CWAs by virtue of its strict verification program.\(^{21}\)

The convention entered into force on 29 April 1997 and has been endorsed by more than 184 countries. Signatory states of CWC are obliged to declare their CW stockpiles and destroy them along with their production facilities. However, India has destroyed her all the stock of CW agents in April 2009 and became the first country in the world believe in the peace. Only 7 Non-Signatory States world-wide have not taken any action on the Convention. They are Angola, Democratic People's Republic of Korea, Egypt, Iraq, Lebanon, Somalia and Syria.

The Convention was also negotiated with the active participation of the global chemical industry, for ensuring industry's on-going cooperation with the CWC's industrial verification regime. According to CWC, industries have to ensure that toxic chemicals are used exclusively for purposes not prohibited by the Convention.\(^{22}\) There are several terms used in the CWC some of them are defined below.

### 5.1 Definitions

**Chemical Weapon**

These are the weapon which contains toxic chemicals or their precursors, which cause death, injury or incapacitation through its chemical action.

**Toxic Chemicals**

Toxic chemicals mean any chemical, which through its chemical action on life process can cause death or temporary incapacitation to human being or animals.
**Precursor**

Precursor means any chemical reactant that takes part at any stage in the production of toxic chemicals.

**Degradation Products**

Any chemical which contains characteristic skeleton of toxic chemical after the chemical reaction like hydrolysis, elimination etc.

**Marker**

A marker may be a precursor, degradation or any reaction product which may be formed in environment under the suitable conditions.

An important feature of the CWC is that it requires a system for verifying compliance. This is implemented by the convention supervising body – The Organization for the Prohibition of Chemical Weapons (OPCW) situated at The Hague, The Netherland.

5.2 The Organization for the Prohibition of Chemical Weapons (OPCW)

The verification program of CWC is pivotal in its implementation, which involves unambiguous identification of CWAs and their related chemicals in samples collected by the OPCW inspectors from different declared and suspected sites, having the concern of homeland security and defence communities, and use of CWAs by terrorist group. OPCW also envisages a very stringent verification program that involves the inspection of different CW agent’s production, storage and industrial sites by inspectors appointed by the OPCW. The OPCW is responsible for verification activities on a regular basis and can conduct challenge inspections. However, challenge inspection of any nation is only possible when that nation is signatory state of the treaty and having undeclared stock of CWAs and objection raised by other nation. Verification involves collection of samples from production, storage and suspected sites by the inspectors appointed by the OPCW. Inspectors perform on-site analysis of collected samples to detect and identify CWC related chemicals.
Where there is ambiguity, the samples are sent to at least two off-site laboratories designated by the OPCW for unambiguous identification of the CWC related chemicals. For the off-site analysis, OPCW is looking for designated laboratories. These laboratories should have instrumental and analytical capability, preparedness, and analytical methods to analyze the samples collected by the OPCW inspectors. The laboratories should be capable to confirm the presence or absence of CWC-related chemicals and other chemicals, in the coded samples sent by the OPCW. The designation of a laboratory is determined through their performance in the Official OPCW Proficiency Tests.

5.3.1 The Official OPCW Proficiency Test

The official OPCW proficiency test (OPT) was started with the objective to simulate sample analysis in order to select laboratories that are capable of performing analysis of chemicals scheduled under the CWC or their degradation products in a wide variety of matrices. It is an inter-laboratory comparison among participating labs from all over the world. The Technical Secretariat of the OPCW conducts OPT twice in a year to check the analytical capability of participating laboratories. To seek and sustain designation, the participating labs have to prove their efficiency by qualifying in three consecutive tests. The participating laboratories must secure minimum two A and one B grade in three consecutive tests to achieve the status of the designated laboratory. Subsequently to sustain the status, a designated laboratory must participate in at least one test per calendar year, and secure A grade. Falling to do so results in suspension or termination of the designation status.

A laboratory may participate in the OPT as a regular participant, whereby the laboratory is given fifteen calendar days to analyze the samples and submit an analysis report to the OPCW. Alternatively, a laboratory may assist in one of two roles, that of the sample preparation laboratory or the evaluating laboratory. The sample preparation laboratory is tasked with formulating the composition of test samples according to a test scenario, performing stability studies to ensure the stability of spiking chemicals in the matrices, preparing the test samples as well as dispatching a set to each of the participating laboratories in addition to two sets each to the evaluating laboratory and the OPCW Laboratory.
On the other hand, the evaluating laboratory is tasked with analyzing the samples using at least two different analytical techniques, at least one of which must be a spectrometric technique, to identify the test chemicals. Thereafter, the evaluating laboratory submits a sample analysis report to the OPCW Laboratory within twenty eight days upon receipt of the samples. Upon receipt of copies of the test reports from participating laboratories, the evaluating laboratory performs a detailed evaluation. A draft preliminary evaluation report is sent to the OPCW Laboratory within twenty eight days upon receipt of the complete set of copies of all participants report. After discussion with the OPCW test coordinator on the draft preliminary evaluation report, a preliminary evaluation report is submitted to the OPCW Laboratory within a week. All participating laboratories that take part in an OPCW proficiency test are awarded a performance rating according to the extent to which the laboratory fulfills the performance.

5.3.1 Challenges of Official Proficiency Test

The official proficiency test involves qualitative analysis of samples. The analysis results should provide sufficient evidences so that presence or absence of CWC related compounds can be confirmed. These tests are blind in nature and the participants do not know the sample composition. The proficiency test does not specify the analytical method but results should be reported by two independent analytical techniques leading to a consistent result. Designation is awarded to the accredited laboratory only. The proficiency test requires detailed reporting of the analysis results. This stringent requirement of sustained performance in OPTs can be fulfilled by (i) developing the spectral database of as many markers of CWAs as possible, because in these blind tests some of the chemicals are spiked for which no spectral data are available world-wide, (ii) a laboratory must excel in sample preparation and analysis of markers of CWAs from various environmental matrices e.g. soil, water, air etc. (iii) laboratory must have efficient synthetic procedure and facility for the synthesizing required reference chemicals.

5.4 Implementation of CWC

The CWC is implemented by the convention supervising body – The Organization for the Prohibition of Chemical Weapons (OPCW). A key objective
of the OPCW is to ensure that the legal prohibitions based on the Convention are comprehensive enough to extend the reach of verification to all scheduled chemicals and new means and methods of production and to prevent their re-emergence. The OPCW also monitors the competence of laboratories and other national agencies tasked with verifying the presence and destruction of chemical weapons and designations of laboratories. These OPCW-designated laboratories are the heart of the OPCW verification regime and of their respective countries.

5.4.1 Verification of CWC

According to CWC guideline, state parties must declare all the storage chemical weapon and destruction sites and state parties are responsible for the destruction of their own chemical weapons. The destruction process is verified by OPCW inspectors through onsite inspection. The verification of CWC involves the sampling and analysis of environmental samples like water, soil etc. to prove the presence or absence of chemicals related to CWAs. For any unresolved ambiguities, these environmental samples may be sent to off-site laboratory.

5.4.2 On-Site Analysis

During the on-site inspections, the OPCW inspectors analyze environmental samples for the presence or absence of the compounds enlisted in the three schedules of CWC or their degradation products. More importantly on-site analysis was conducted at an inspection site or an incident site for rapid responses to CWA incidents. On-site analysis can also be critical for verifying declared industrial chemicals and OPCW challenge inspections in which transporting samples away from the premises is discouraged due to trade secrets or regulatory requirements.28

5.4.3 Off-Site Analysis

In the event of off-site analysis, authentic samples will pass from the inspection site through the OPCW Laboratory to designated laboratories. A designated laboratory receives for each sample, three non-indicated vials containing sample, spiked control, and blank. The received sample under goes sample preparation and their analysis by different chromatographic and spectrometric techniques. Each laboratory actually requires its own analysis strategy for the analysis of sample. Unambiguous identification of a chemical is obtained if at least two analytical, preferably spectrometric, techniques give consistent results. Off- site
analysis results are assessed by the OPCW laboratory for correct analysis of control samples and blanks, for conforming to relevant reporting criteria and for consistency of results between designated laboratories. The testing report is prepared according to the OPCW reporting criteria and sent to the OPCW within 15 days.

5.5 Chemicals Related to the Chemicals Weapons Convention

The CWC related chemicals are annexed in three schedules of treaty, which covers several thousands of chemicals. The division into the schedules is based on the degree of risk the chemicals pose to the object and extend of their commercial application.\(^\text{29}\)

5.5.1 Schedule 1

These are the chemicals which can either be used as chemical weapons themselves or used in the manufacture of chemical weapons and which have no, or very limited, uses outside of chemical warfare. These chemicals may be produced or used for research, medical, pharmaceutical or chemical weapon defence testing (called "protective testing" in the treaty) purposes but production above 100 grams per year must be declared to the OPCW.

The following criteria shall be taken into account in considering whether a toxic chemical or precursor should be included in Schedule 1:

- It has been developed, produced, stockpiled or used as a chemical weapon.
- It possesses a chemical structure closely related to that of other toxic chemicals listed in Schedule 1, and has, or can be expected to have, comparable properties;
- It possesses such lethal or incapacitating toxicity as well as other properties that would enable it to be used as a chemical weapon;
- It may be used as a precursor in the final single technological stage of production of a toxic chemical listed in Schedule 1
5.5.2 List of the Schedule 1 Chemicals

A. Toxic chemicals:

(1) O-Alkyl (<C10, incl. cycloalkyl) alkyl Me, Et, n-Pr or i-Pr)-phosphonofluoridates
   e.g. Sarin: O-Isopropyl methylphosphonofluoridate
   Soman: O-Pinacolyl methylphosphonofluoridate

(2) O-Alkyl (<C10, incl. cycloalkyl) N,N-dialkyl Me, Et, n-Pr or i-Pr) phosphoramidocyanidates
   e.g. Tabun: O-Ethyl N,N-dimethyl phosphoramidocyanidate

(3) O-Alkyl (H or <C10, incl. cycloalkyl) S-2-dialkyl (Me, Et, n-Pr or i-Pr)-aminoethyl alkyl
   (Me, Et, n-Pr or i-Pr) phosphonothiolates and corresponding alkylated or protonated salts
   e.g. VX: O-Ethyl S-2-diisopropylaminoethyl methyl phosphonothiolate

(4) Sulfur mustards:
   2-Chloroethylchloromethylsulfide
   Mustard gas: Bis(2-chloroethyl)sulfide
   Bis(2-chloroethylthio)methane
   Sesquimustard: 1,2-Bis(2-chloroethylthio)ethane
   1,3-Bis(2-chloroethylthio)-n-propane
   1,4-Bis(2-chloroethylthio)-n-butane
   1,5-Bis(2-chloroethylthio)-n-pentane
   Bis(2-chloroethylthiomethyl)ether
   O-Mustard: Bis(2-chloroethylthioethyl)ether

(5) Lewisite:
   Lewisite 1: 2-Chlorovinylidichloroarsine
   Lewisite 2: Bis(2-chlorovinyl)chloroarsine
   Lewisite 3: Tris(2-chlorovinyl)arsine

(6) Nitrogen mustards:
   HN1: Bis(2-chloroethyl)ethylamine
   HN2: Bis(2-chloroethyl)methylamine
   HN3: Tris(2-chloroethyl)amine
(7) Saxitoxin

(Saxitoxin)

(8) Ricin

B. Precursors:

(9) Alkyl (Me, Et, n-Pr or i-Pr) phosphonyldifluorides
e.g. DF: Methylphosphonyldifluoride

(10) O-Alkyl (H or <C10, incl. cycloalkyl) O-2-dialkyl
(Me, Et, n-Pr or i-Pr)-aminoethyl alkyl
(Me, Et, n-Pr or i-Pr) phosphonites and corresponding alkylated or
protonated salts
e.g. QL: O-Ethyl O-2-diisopropylaminoethyl methylphosphonite

(11) Chlorosarin: O-Isopropyl methylphosphonochloridate

(12) Chlorosoman: O-Pinacolyl methylphosphonochloridate

5.5.3 Schedule 2

These are the chemicals that can either be used as chemical weapons themselves
or used in the manufacture of chemical weapons but that have small-scale
applications outside of chemical warfare and so can be legitimately
manufactured in small quantities. An example is thiodiglycol, which can be used
in the manufacture of mustard agents but is also used as a solvent in inks.
Manufacture must be declared as their production is subject to declaration to
the OPCW, and they may not be exported to countries that are not party to the
Convention

The following criteria shall be taken into account in considering whether a toxic
chemical should be included in Schedule 2:
- It poses a significant risk to the object and purpose of this Convention because it possesses such lethal or incapacitating toxicity as well as other properties that could enable it to be used as a chemical weapon;

- It may be used as a precursor in one of the chemical reactions at the final stage of formation of a chemical listed in Schedule 1 or Schedule 2.

5.5.4 List of the Schedule 2 Chemicals

A. Toxic chemicals:

(1) Amiton: O,O-Diethyl S-[2-(diethylamino)ethyl] phosphorothiolate and corresponding alkylated or protonated salts

![Amiton-VG]

(2) PFIB: 1,1,3,3,3-Pentafluoro-2-(trifluoromethyl)-1-propene

![PFIB]

(3) BZ: 3-Quinuclidinyl benzilate

B. Precursors:

(4) Chemicals, except for those listed in Schedule 1, containing a phosphorus atom to which is bonded one methyl, ethyl, isopropyl or propyl group only.

  e.g. Methylphosphonyl dichloride, Dimethyl methylphosphonate

(5) N,N-Dialkyl (Me, Et, n-Pr or i-Pr) phosphoramidic dihalides

(6) Dialkyl (Me, Et, n-Pr or i-Pr) N,N-dialkyl (Me, Et, n-Pr or i-Pr)-phosphoramidates

(7) Arsenic trichloride

(8) 2,2-Diphenyl-2-hydroxyacetic acid

(9) Quinuclidin-3-ol
(10) N,N-Dialkyl (Me, Et, n-Pr or i-Pr) aminoethyl-2-chlorides and corresponding protonated salts
(11) N,N-Dialkyl (Me, Et, n-Pr or i-Pr) aminoethane-2-ols and corresponding protonated salts
(12) N,N-Dialkyl (Me, Et, n-Pr or i-Pr) aminoethane-2-thiols and corresponding protonated salts
(13) Thiodiglycol: Bis(2-hydroxyethyl)sulfide
(14) Pinacolyl alcohol: 3,3-Dimethylbutan-2-ol

5.5.5 Schedule 3

These are the chemicals which can either be used as toxic chemical weapons themselves or used in the manufacture of chemical weapons but which also have legitimate large-scale industrial uses. Plants which manufacture of more than 30 tonnes per year must be declared and can be inspected, and there are restrictions on export to countries which are not CWC signatories. Examples of these substances are phosgene, which has been used as a chemical weapon but which is also a precursor in the manufacture of many legitimate organic compounds similarly triethanolamine, used in the manufacture of nitrogen mustard but also commonly used in toiletries and detergents.

The following criteria shall be taken into account in considering whether a toxic chemical should be included in Schedule 3:

- It has been produced, stockpiled or used as a chemical weapon.
- It poses otherwise a risk to the object and purpose of this Convention because it possesses such lethal or incapacitating toxicity as well as other properties that might enable it to be used as a chemical weapon.
- It may be produced in large commercial quantities for purposes not prohibited under this Convention.
5.5.6 List of the Schedule 3 Chemicals

A. Toxic chemicals:
   (1) Phosgene: Carbonyl dichloride
   (2) Cyanogen chloride
   (3) Hydrogen cyanide
   (4) Chloropicrin: Trichloronitromethane

B. Precursors:
   (5) Phosphorus oxychloride
   (6) Phosphorus trichloride
   (7) Phosphorus pentachloride
   (8) Trimethyl phosphite
   (9) Triethyl phosphite
   (10) Dimethyl phosphite
   (11) Diethyl phosphite
   (12) Sulfur monochloride
   (13) Sulfur dichloride
   (14) Thionyl chloride
   (15) Ethyldiethanolamine
   (16) Methyldiethanolamine
   (17) Triethanolamine

The chemicals contained in the above three schedules are mainly organic chemicals with different chemical and physical properties, such as neutral chemicals, acids, bases, volatiles, and non-volatile chemicals, where the elements phosphorous, fluorine, sulfur, chlorine, nitrogen, and oxygen occur frequently.

6. General methods for the synthesis of precursor, degradation product and CWAs

The most popular reactions of organophosphorus compounds are described below. These synthetic methods can be used for the synthesis of CWC related compounds.
6.1 Synthesis of precursors

These are the chemicals required for the synthesis of various CWAs related compounds. There synthesis is as follows

6.1.1 Michaelis-Arbusov Reaction

In this reaction, trialkyl phosphites react with alkyl or acyl halides to produce corresponding phosphonate esters. This reaction is one of the most versatile methods for the formation of P-C bonds and is well known as Michaelis-Arbusov reaction. General scheme of this reaction are as follows:

Scheme 1: Michaelis-Arbusov reaction

6.1.2 Kinnear and Perren reaction

In this reaction, alkyl halide reacts readily with phosphorus trichloride and aluminium chloride to give complexes which on treatment with cold water gives good yields of alkyl phosphonic dichlorides.

Scheme 2: Kinnear and Perren reaction
6.1.3 Michaelis-Becker-Nylen reaction

In this reaction dialkylphosphites are treated with sodium metal in equimolar ratio and sodium salt of dialkylphosphite thus obtained is allowed to react with alkyl halides to afford products with P-C bond. It may be regarded as an extension of Michaelis-Becker reaction.\(^{33-34}\)

It has been found that reactivity order of phosphites are:

\[
(\text{CH}_3\text{O})_2\text{P}-\text{OH} > (\text{CH}_3\text{CH}_2\text{O})_2\text{P}-\text{OH} > (\text{CH}_3\text{CH}_2\text{CH}_2\text{O})_2\text{P}-\text{OH}
\]

and

\[
\text{R-I} > \text{R-Br} > \text{R-Cl}
\]

for alkyl halides.

\[\text{Scheme 3: Michaelis-Becker-Nylen reaction}\]

6.1.4 Modified Kinnear and Perren reaction

In another method, modified form of Kinnear Perren’s reaction, was also used for the synthesis of alkyl phosphonothioic dichloride in one step by using thiourea.\(^{35}\)

\[
\text{R-Cl} + \text{PCL}_3 + \text{Al}_2\text{Cl}_6 \rightarrow \left[\text{R-PH-Cl}_3\right]^4 \left[\text{Al}_2\text{Cl}_7\right]^{-}
\]

\[\text{(i) Thiourea}\]
\[\text{(ii) Water}\]

\[\text{Scheme 4: Modified Kinnear and Perren reaction}\]
6.1.5 Synthesis of \( \text{P=S} \) linkage

Compounds containing \( \text{P=S} \) bond(s) are extremely important as many of CWC related compounds. Alkyl phosphonic dichloride is converted their corresponding alkyl phosphonic dichloride by reacting with phosphrous pentasulphide.\(^{36}\)

![Scheme 5: Synthesis of alkyl thiophosphonic dichloride](image)

6.2 Synthèses of Degradation Products

These are the chemical generated after decomposition of the CWAs in the environment or by using suitable decontamination reagents. Their synthesis are as follows

6.2.1 Synthesis of \( \text{O,O'} \)-dialkyl alkylphosphonates and \( \text{O-alkyl alkylphosphonic acids} \)

Surface mediated synthesis of \( \text{O,O'} \)-dialkyl alkylphosphonates and \( \text{O-alkyl alkylphosphonic acid} \) was synthesized using N,N'-Dicyclohexylcarbodiimide (DCC) as a catalyst. The developed method was used for the synthesis of various degradation products of nerve agents.\(^{37}\)

![Scheme 6: Synthesis of \( \text{O,O'} \)-dialkyl alkylphosphonates and \( \text{O'-alkyl hydrogen alkylphosphonates using DCC.} \)

6.2.2 Synthesis of \( \text{O,O'} \)-dialkyl alkylphosphonates and \( \text{O-alkyl hydrogen alkylphosphonates on Silica Plate} \)

In this technique, the reactants and catalysts are co-spotted on silica thin layer chromatography (TLC) followed by microwave irradiation or heating. The desired compound are scratched from the TLC plate followed by washed with a suitable solvent to obtained the desired compound.\(^{38}\)
Scheme 7: Synthesis of $O,O'$-dialkyl alkylphosphonates and $O$-alkyl hydrogen alkylphosphonates on TLC.

6.2.3 Synthesis of $O,O'$-dialkyl $N,N$-dialkyl phosphoramidates

In this study synthesis of dialkyl chlorophosphates from dialkylphosphites using Polymeric solid supported chlorinated reagent (N,N-dichloro poly(styrene-co-divinyl benzene)sulfonamide.\(^{39}\) Which was further used for the synthesis of $O,O'$-dialkyl $N,N$-dialkyl phosphoramidates by the treatment with corresponding secondary amine.

Scheme 8: Synthesis of $O,O'$-dialkyl $N,N$-dialkyl phosphoramidates

6.2.4 Solid Supported Synthesis of CWC related phosphoramidates

In this study Al$_2$O$_3$-KOH supported condensation of phosphoramidic dichlorides with alcohols yielded the CWC related dialkyl $N,N$-dialkylphosphoramidates.\(^{40}\)

Scheme 9: Synthesis of $O,O'$-dialkyl $N,N$-dialkyl phosphoramidates
6.3 Synthesis of Different Types Chemical Warfare Agents

Different types of chemical warfare agents are can be synthesized based on the following synthetic procedures.\(^{41}\)

6.3.1 Sulphur mustard

Sulphur mustard (Mustard gas) can be prepared in a two-step process starting with the formation of bis(hydroxyethyl) sulphide. This hydroxyethyl sulphide intermediate is easily prepared by reacting water solutions of sodium sulphide and ethylene chlorohydrine. The resulting hydroxyethyl sulphide intermediate is then converted into mustard gas by the addition of concentrated hydrochloric acid. After the addition of concentrated hydrochloric acid, the mustard gas will separate as viscous oil. The mustard gas is then separated by separating funnel, followed by vacuum distilled to get distilled mustard gas.

\[
\text{Scheme 10: Synthesis of Sulphur Mustard}
\]

6.3.2 Lewisite

Lewisite can be prepared by the addition of acetylene gas to a mixture of Arsenic trichloride (AsCl\(_3\)) and concentrated hydrochloric acid containing a certain amount of mercuric chloride (HgCl\(_2\)) as a catalyst at 50\(^\circ\)C. After the addition of acetylene gas the reaction mixture is stirred and refluxed for another 30 minutes. The resulting lewisite layer is then removed by separating funnel, followed by vacuum distilled to get the distilled lewisite.

\[
\text{Scheme 11: Synthesis of Tris (2-chloroethenyl) Arsine}
\]
6.3.3 Nitrogen Mustard (HN3)

Nitrogen Mustard (HN3) can be easily prepared by reacting triethanolamine with thionyl chloride in chloroform. The addition of thionyl chloride should be carried out at room temperature followed by refluxing at 70°C for 2 hour. After that cool the reaction mixture and remove the excess of solvent to get HN3 hydrochloride salt. Which is further treated with alkaline solution to obtain the desired nitrogen mustard.

\[
\begin{array}{c}
\text{HO} & \text{N} & \text{OH} \\
\text{Cl} & & \\
\text{CHCl}_3, 70^\circ C & \text{HN} & \text{Cl} \\
\text{Cl} & & \text{NaHCO}_3 & \text{N} & \text{Cl} \\
\end{array}
\]

Scheme 12: Synthesis of 2, 2', 2''-trichlorotriethylamine

6.3.4 Chloropicrin

Chloropicrin (Chocking agent) is easily prepared by reacting sodium hypochlorite with nitromethane for four hours at room temperature. Then after insoluble chloropicrin layer is obtained, which is separated by separating funnel.

\[
\text{NaOCl} + \text{H}_3\text{C}N+\text{O}^- \rightarrow \text{Cl}\text{C-O-N}^+\text{O}^-
\]

Scheme 13: Synthesis of Chloropicrine

6.3.5 Hydrogen Cyanide

Hydrogen cyanide (Blood agent) is readily prepared by reacting potassium cyanide with dilute sulphuric acid.

\[
\text{KCN} \xrightarrow{\text{H}_2\text{SO}_4} \text{HCN}
\]

Scheme 14: Synthesis of Hydrogen cyanide
6.3.6 Nerve agent

These agents are the extremely toxic agent among other CWAs. Sarin, soman, cyclo sarin belongs to the organophosphorous fluoridate. They are synthesized as follows presented with example of sarin. This chemical \( O \)-isopopyl methylphosphonoflouridate (sarin) can be prepared by the treatment of trimethylphosphite with methyl iodide to get dimethyl methylphosphonate. Then it is further treated with thionyl chloride followed by isopropanol to obtained o-isopropyl methyphosphonochloridate. Finally, it is treated with sodium fluoride to get the desired compound i.e. Sarin.

Scheme 15: Synthesis of Sarin

7. Characterisation of organophosphorus compounds

The structures of organophosphorus compounds are very similar to other organic compounds. It has been greatly accelerated by the development of general spectroscopic methods, especially Fourier Transform Infrared spectroscopy (FTIR), Nuclear Magnetic Resonance (NMR) and Mass spectrometry (MS).
Chapter-1

7.1 Infrared spectroscopy

Some of the characteristic IR absorption frequencies of organophosphorus compounds are given below in the following table.\textsuperscript{42}

Table 7: Infrared absorption frequencies of organophosphorus compounds

<table>
<thead>
<tr>
<th>Sl. No</th>
<th>Functional Group</th>
<th>IR absorption frequencies</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>P-F stretching</td>
<td>900-720 cm(^{-1})</td>
</tr>
<tr>
<td>2</td>
<td>P-Cl stretching</td>
<td>580-440 cm(^{-1})</td>
</tr>
<tr>
<td>3</td>
<td>P-Br stretching</td>
<td>485-400 cm(^{-1})</td>
</tr>
<tr>
<td>4</td>
<td>P-C stretching</td>
<td>770-650 cm(^{-1})</td>
</tr>
<tr>
<td>5</td>
<td>P-N stretching</td>
<td>750-680 cm(^{-1})</td>
</tr>
<tr>
<td>6</td>
<td>P-O stretching</td>
<td>1300-1200 cm(^{-1})</td>
</tr>
<tr>
<td>7</td>
<td>P-H stretching</td>
<td>2270-2450 cm(^{-1})</td>
</tr>
<tr>
<td>8</td>
<td>P=O stretching</td>
<td>1350 – 1250 cm(^{-1})</td>
</tr>
<tr>
<td>9</td>
<td>P=S stretching</td>
<td>750-600 cm(^{-1})</td>
</tr>
<tr>
<td>10</td>
<td>P=N stretching</td>
<td>1300-1100 cm(^{-1})</td>
</tr>
<tr>
<td>11</td>
<td>P-O-P linkages</td>
<td>970-870 cm(^{-1})</td>
</tr>
<tr>
<td>12</td>
<td>P-O-C (aliphatic)</td>
<td>1050-980 cm(^{-1})</td>
</tr>
<tr>
<td>13</td>
<td>P-O-C (aromatic)</td>
<td>1240 – 1190 cm(^{-1})</td>
</tr>
<tr>
<td>14</td>
<td>P-CH(_3) group</td>
<td>1320 – 1280 cm(^{-1})</td>
</tr>
<tr>
<td>15</td>
<td>P-S-C linkage</td>
<td>600 – 500 cm(^{-1})</td>
</tr>
</tbody>
</table>

7.2 NMR spectroscopy

Nuclear magnetic resonance (NMR) spectroscopy is a valuable analytical technique for the identification of CWC related compounds. The presence of hetero nuclei such as \(^{31}\)P and \(^{19}\)F in the nerve agents leads to diagnostic splitting patterns and coupling constants due to \(^1\)H-\(^{31}\)P and \(^1\)H-\(^{19}\)F spin-spin coupling. Specific hetero nuclear experiments such as \(^{31}\)P NMR is recommended in screening CWC related organophosphorus compounds in complex matrices. Characteristic chemical shifts of compounds containing a phosphorus-carbon bond and splitting due to phosphorus-fluorine spin-spin coupling can be used to screen for the presence of nerve agents. The chemical shift values of \(^{31}\)P NMR
range between +225 to -450 ppm for all type of compounds. Some of the important class of organophosphorus compounds are given below (table 2) with their absorption range (reference standard is H$_3$PO$_4$).

Table 8: Chemical shift Organophosphous Compounds

<table>
<thead>
<tr>
<th>Sl.No.</th>
<th>Name of the Compounds</th>
<th>$^{31}$P chemical shift NMR range</th>
<th>$^{19}$F chemical shift range</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>$O$-alkyl $N,N$-dialkylphosphoramidocydanides</td>
<td>-5 to -17 ppm (s)</td>
<td>-</td>
</tr>
<tr>
<td>2.</td>
<td>Alkylphosphoramic difluorides</td>
<td>10 to -16 ppm (t)*</td>
<td>-73 to -82 ppm (d)</td>
</tr>
<tr>
<td>2.</td>
<td>Alkylphosphoramic dichlorides</td>
<td>15 to -20 ppm (s)</td>
<td>-</td>
</tr>
<tr>
<td>3.</td>
<td>$O$-alkyl alkylphosphorimidates</td>
<td>7 to 20 ppm (s)</td>
<td>-</td>
</tr>
<tr>
<td>4.</td>
<td>Alkylphosphonic difluorides</td>
<td>16 to 45 ppm (t)</td>
<td>-62 to -79 ppm (d)</td>
</tr>
<tr>
<td>5.</td>
<td>Alkylphosphonic dichlorides</td>
<td>63 to 42 ppm (s)</td>
<td>-</td>
</tr>
<tr>
<td>6.</td>
<td>Alkyl phosphonic diacids</td>
<td>20 to 37 ppm (s)</td>
<td>-</td>
</tr>
<tr>
<td>7.</td>
<td>$O,O$-dialkyl alkylphosphonates</td>
<td>26 to 40 ppm (s)</td>
<td>-</td>
</tr>
<tr>
<td>8.</td>
<td>$O$-alkyl alkylphosphonates (monoacids)</td>
<td>24 to 40 ppm (s)</td>
<td>-</td>
</tr>
<tr>
<td>9.</td>
<td>$O$-alkyl alkylphosphono-fluoridates</td>
<td>20 to 40 ppm</td>
<td>-50 to -70 ppm</td>
</tr>
<tr>
<td>10.</td>
<td>$O$-alkyl alkylphosphono-chloridates</td>
<td>37 to 42 ppm (s)</td>
<td>-</td>
</tr>
<tr>
<td>11.</td>
<td>Alkylthiophosphonic difluorides</td>
<td>93 to 117 ppm (t)</td>
<td>-24 to -62 ppm (d)</td>
</tr>
<tr>
<td>12.</td>
<td>Alkylthiophosphonic dichlorides</td>
<td>84 to 104 ppm (s)</td>
<td>-</td>
</tr>
<tr>
<td>13.</td>
<td>Alkylthiophosphonic acids</td>
<td>72 to 99 ppm (s)</td>
<td>-</td>
</tr>
<tr>
<td>14.</td>
<td>$O,O$-dialkyl alkylthiophosphonates</td>
<td>89 to 107 ppm (s)</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>Chemical Structure</td>
<td>δ ppm (Assignments)</td>
<td></td>
</tr>
<tr>
<td>---</td>
<td>-------------------</td>
<td>---------------------</td>
<td></td>
</tr>
<tr>
<td>15.</td>
<td>$O$-alkyl alkylthiophosphono-fluoridates</td>
<td>94 to 110 ppm (d) -35 to -46 ppm (d)</td>
<td></td>
</tr>
<tr>
<td>16.</td>
<td>$O$-alkyl alkylthiophosphonates (monoacids)</td>
<td>90 to 105 ppm (s) -</td>
<td></td>
</tr>
<tr>
<td>17.</td>
<td>Dialkyl phosphite</td>
<td>6 to 13 ppm (s) -</td>
<td></td>
</tr>
<tr>
<td>18.</td>
<td>Trialkyl phosphite</td>
<td>130 to 150 ppm (s) -</td>
<td></td>
</tr>
<tr>
<td>20.</td>
<td>Alkyl S-2-(dialkylamino)ethyl alkylphosphonothiolate</td>
<td>52 to 67 ppm (s) -</td>
<td></td>
</tr>
</tbody>
</table>

### 7.3 Mass spectrometry

Mass spectrometry (MS) is the important method of choice for the detection and characterization of chemical warfare agents, their precursors, degradation products and related compounds.

The majority of MS data have been obtained under electron impact (EI) ionization conditions. However many of the convention related chemicals (CRCs) do not provide molecular ion information under EI-MS. This hinders confirmation of these analytes and makes identification difficult. For this reason, considerable effort has been devoted to the use of chemical ionization (CI) as a complementary ionization technique. This milder form of ionization generally affords molecular ion information for the CRCs and has been used extensively for the identification of compounds.

### 9. Safety and disposal

Laboratory safety is the first step in proper laboratory techniques. Chemical warfare agents are extremely hazardous and lethal compounds. They should only be synthesized in well-equipped laboratories by trained personals. Safety and standard operating procedures must be developed and approved before executing the work related to chemical warfare agents. Chemical warfare agents should be synthesized only in that laboratory which should have scrubber system or efficient fume hood. Personnel handling chemical warfare agents should wear protective gears (safety goggles, lab coats, full face mask, and nitrile rubber gloves) boots, and therapeutic devices within easy reach. Electronic monitored system should be used to immediately alert personals to the slightest presence of
toxic agents. Sufficient decontamination should be available to destroy the
effluents generated from the synthesis of chemical warfare agents.
Decontaminated chemical warfare agents must be disposed of in an
environmentally approved method according to local legislation. Currently used
decontaminants include nucleophile/base-amine mixtures and bleach
formulations.\textsuperscript{43-44} Nucleophile/base-amine decontaminants are the mixtures of
bases (alkali metal hydroxides, limes, potash, etc) and nucleophiles (phenoxides,
glycols) with amines like diethylenetriamine, ammonia and monoethanolamine.
Most popular and widely used reactive decontaminant is ‘Decontamination
Solution-2’ (DS-2) which is very effective in deactivating the CWAs. Bleach
formulations including microemulsions are made of positive halogen (mostly
chlorine) carrying compounds like calcium hypochlorite, sodium hypochlorite,
chloramines (dichloroisocyanuric acid, dichloramine, chloramine-T, Alkali metal
hypochlorites etc).

10. **Objective of the work**
The use of CWAs in war or by terrorist group is the first choice as a weapon of
mass destruction, because these can be easily synthesized by simple chemical
reactions and they possess extreme toxicity. After world war II, CWA’s have
been used occasionally both in war and public places such as in the Iraq-Iran war
and Japan underground rail station attacks. Recently, attack of nerve agent in
Syria (2013) alert the international community to take necessary steps for the
detection, protection and decontamination of CWAs. Experts believe that
terrorist groups may acquire the insecure stockpiles of chemical weapon which
becomes an important issue for every nation in the world.

An off-site analysis of collected samples is a sensitive matter as it pertains to
proving compliance / non-compliance of the CWC. Therefore, the verification
analysis of CWC necessitates adoption of analytical techniques, sample
preparation and final confirmation of the analytes only after synthesizing the
reference chemicals.

A large number of reference chemicals and their spectra are required to enrich
the on-line spectral data base and to sustain the designation in proficiency tests.
Since majority of the chemicals, as enlisted in the CWC are not commercially
available, their synthesis becomes an inevitable activity during the verification
analysis. The verification analysis of CWA’s require detection and identification of not only the toxic schedule 1 chemicals, but also their specific non-toxic ‘markers’ which are present as either by-products or produced by a chemical reaction after spillage / spread of nerve agents. These non-toxic markers of nerve agents are covered in the schedule 2B4 category of CWC and encompass several hundred thousands of chemicals. There are various methods reported in the literature for the synthesis of reference chemicals for the verification of chemical warfare agents. Since, proficiency test is a time bound activity and the list of schedule compounds is exhaustive. So, there is dire need to develop efficient and new synthetic strategies which can be used for convenient synthetic procedures for the synthesis of reference chemicals for the verification of chemical warfare agents. Thus, considering the limited availability of spectral database of reference chemicals and its requirements for national and international program of verification of CWC, their study was undertaken with following objectives.

1. To develop simple and efficient synthetic procedures for the rapid synthesis of $O,O'$-dialkyl alkylphosphonates (scheduled 2B4) by using polymer supported and magnetic nanocatalyst.

2. To develop a new, synthetic procedure for the synthesis of highly toxic nerve agent simulant i.e dialkyl fluoro phosphates by using microflow technique and by using inorganic reagents.

3. To develop a new rapid and efficient synthetic procedures for preparation of $O,O'$-dialkyl $N,N$-dialkylphosphoramidates ( marker of tabun nerve agents) by using inorganic reagents.

11. **Significance of Present study**

A large number of CWC related chemicals and their spectra are required to improve the in-house spectral library and to sustain the designation in official proficiency tests. Majority of the chemicals as enlisted in the CWC under the heading of three schedules and their precursors as well as degradation product are not commercially available and their synthesis becomes an inevitable activity during the verification analysis. Since reported methods have several drawbacks such as use of hazardous solvent, long reaction time, require a strong base, high temperature, tedious workup procedure and low yield of the products.
The present study has been carried out keeping in view that the improved synthetic methods will be helpful for the synthesis of various reference chemicals by using different functionalizing polymer and magnetic supported nanocatalyst with easy work up procedure. The highly toxic chemical can be synthesized in a closed system (microflow system) without direct exposur during its synthesis. Nerve agents simulants were synthesized having smart work-up procedure. At present, researchers are focusing on the development of special methodologies on detection, protection and decontamination of these nerve agents. Thus the chemistry of CWC related compounds is putting the challenge on synthetic chemists and demand to have the expertise. Thus the objective of this work was to develop newer efficient and convenient synthetic methods for the synthesis of reference compounds pertaining to detection and identification. The spectral data base generated by this study will be useful for the reproducible synthesis and identification of chemical warfare agents
12. Reference


11. J.R. Cashman, Emergency Response to Chemical and Biological Agents, CRC


