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REVIEW ARTICLE

BIOLOGICAL WEAPONS – HISTORY AND PRESENT

Ales Macela [1], Vanda Bostikova [2], Klara Kubelkova [3] ✉

[1] Military Faculty of Medicine, University of Defense, Třebešská 1575, 500 01 Hradec Králové, Czech Republic
[2] Department of Epidemiology, Military Faculty of Medicine, University of Defense, Třebešská 1575, 500 01 Hradec Králové, Czech Republic
[3] Department of Molecular Pathology and Biology, Military Faculty of Medicine, University of Defense, Třebešská 1575, 500 01 Hradec Králové, Czech Republic

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Summary

In 2025, we commemorated the 50th anniversary of the ratification of the Convention on the Prohibition of the Development, Production and Stockpiling of Bacteriological (Biological) and Toxin Weapons and on their Destruction (Biological Weapons Convention, BWC). By 2025, 189 countries had signed the treaty, including the four original signatory states. However, research on microorganisms or their products – toxins – that can be misused for military or terrorist purposes is still being carried out in military facilities in many countries. For this reason, constant attention should be paid to the issues related to the individual Articles of the convention. This article, taking advantage of this anniversary, seeks to provide a precise definition of biological weapons, a brief historical overview of the use of biological agents in armed conflict, and a brief description of the efforts to prohibit the use of microorganisms or their products as weapons of mass destruction, especially against human targets. In its concluding remarks, given the overlap between biomedical and military research on the use of biological agents. The text addresses the still not completely resolved issue of the misuse of biological agents, referring to the long and layered history of their use for nefarious purposes, contributes to distinguish between defensive and offensive biological research, and draws attention to the complexity of issues related to the biological security of world´s states.

Key words: Biological Weapons Convention; biological agents; biological warfare; history

Introduction

The Biological Weapons Convention (BWC) celebrated its 50th anniversary in 2025. The purpose of this international treaty was to prohibit the development, production, and stockpiling of biological and toxin weapons. The convention entered into force on March 26, 1975, and 189 states have ratified it (1,2). Despite this convention, however, no general consensus has been reached against the development of biological weapons (BW), which are now illegal (3,4). An authoritative definition of BW endorsed by the World Health Organization in a report which narrowly pre-dated the adoption of the BTWC and, presumably, was taken into account by negotiators, describes weaponized biological agents as including "those that depend for their effects on multiplication within

✉ University of Defense, Military Faculty of Medicine, Department of Molecular Pathology and Biology, Třebešská 1575, 500 01 Hradec Králové, Czech Republic
klara.kubelkova@unob.cz

the target organism, and are intended for use in war to cause decease in man, animals or plants" (Report of a WHO Group of Consultants, "Health Aspects of Chemical and Biological Weapons", Geneva, 1970, p. 12). From a historical perspective of BW, we may paraphrase one of the Bond films, in which Sean Connery solves a case involving the smuggling of diamonds mined in South Africa, "*Diamonds Are Forever*," it can be said with complete certainty that *biological weapons are eternal*. They came into being together with the genetic differentiation and geographical spread of various microbial, plant, and animal species, including humans. Individual biological taxa use them to protect their own territory or their own interests, only they are not called biological weapons. The reason for their emergence is the fundamental characteristics of living matter, namely its ability to constantly generate diversity and occupy every space available to it, including space already occupied by other living matter. For nature, biological weapons are a means of defense; it was humans who began to use them as a tool of coercion. Bacteria produce toxins (5), molds produce antibiotics (6,7), fungi produce several types of mycotoxins (8,9), all of which serve either for defense or for obtaining food. If we start with plants, their poisons have a primarily defensive role. Plant alkaloids, such as citisine from plants of the Fabaceae family *(Fabaceae*) (10) or coniine from spotted hemlock (*Conium maculatum*) (11), taste bitter and are highly toxic. Many essential oils have an antibacterial effect, killing pathogenic microorganisms, and thanks to their distinctive scents, they serve as protection against herbivores or to attract pollinators. Enzymes produced by plants can also have a defensive or regulatory function (12). Animals also produce a number of poisons. Hymenoptera (bees, wasps, hornets) (13), snakes, scorpions, and some amphibians, such as the common toad or the so-called arrow frogs in the Amazon, have very effective biological weapons (14). The dominant functions of animals' biological weapons are either protection against predators or use by predators to obtain food. And this brings us very close to the purpose for which humans have used, and perhaps still plan to use, biological weapons.

Biological weapons – what does it mean

A weapon, in its most general sense, is any device that allows force to be applied to another object or organism for the purpose of defense, attack, or gain an advantage. In the human world, the term usually refers to artificially made tools intended for combat (knives, firearms, chemical agents, etc.), but biologically it is a much broader category. In biology and technology, the term can be divided according to function into offensive, defensive, deterrent, and specialized weapons, evolutionarily adapted to a very specific purpose. It can be morphological, chemical, behavioral, or technological.

In animals, weapons are considered to be any anatomical or physiological structures that serve for defense or attack. The shell of a turtle, the chemical sprays of some beetles, even mimicry and aposematic coloration (a conspicuous warning coloration that signals to predators that its bearer is poisonous or inedible), are all weapons of passive defense. The poisonous teeth of snakes, the claws of felines or the electric organs of the electric eel are defined as offensive weapons. There are also other "biological" weapons for obtaining food, such as spider webs as a passive trap that immobilizes prey, the shape of the head of hammerheads for fixing prey or a jaw catapult fired at enormous speed. Weapons in animals arise where they bring an evolutionary advantage. This lies in the use of one of the basic properties of living matter, which is to occupy any space that it has available, even space that is already occupied by living matter such as see intracellular and other parasites.

Humans themselves do not have any organs that produce molecules harmful to other animal species. In order to survive as a biological species, they had to use another weapon, the brain, and began to create. Taught by nature, they defined a defense strategy using technical and biological means. The term biological means, i.e., weapons, included everything that could harm other humans, livestock, or crops. This is essentially the content of all definitions related to biological weapons. The most frequently cited definition is that "biological and toxin weapons are either microorganisms, such as viruses, bacteria, or fungi, or toxic substances produced by living organisms, which are produced and released intentionally to cause illness and death in humans, animals, or plants." This is not an official WHO definition, but this characterization is given on their website (15). Perhaps a more accurate concept of BW is that of a technical weapon system consisting of the biological agent itself, often referred to as B agent for short, and the means of delivery to the target and its targeted dissemination (missile warheads, aerial bombs, spraying and dispersal devices) (16). For the sake of simplicity, we will not deal with the technical means, but will focus exclusively on biological agents and their possible misuse, and we will refer to them as biological weapons (hereinafter BW) in accordance with the common definition.

We must emphasize, in accordance with BWC Article I, that a microorganism or its toxic product can only be considered a biological weapon when it is used for evil. Microorganisms are not biological weapons in and of themselves. For example, anthrax spores in soil are not a “biological weapon”; it is a natural pathogen. It only becomes a biological weapon when someone isolates it and prepares it for an attack.

Historical data on the use of biological agents as potential weapons

The earliest documented incident of the intended use of BW is probably recorded in Hittite religious texts from 1500–1200 BC, in which "victims of tularemia" were driven into enemy countries, in the case described into the country of Azarwa (Arzawa, an ancient coalition of principalities in western Anatolia), causing a "plague" epidemic that affected the entire region (17). The causative agent of tularemia, also known as rabbit fever, is the bacterium *Francisella tularensis*. Although *Francisella tularensis*, subsp. *tularensis* is considered a biologically exploitable agent for military purposes, it could not have been the cause of the epidemic, as there is no known human-to-human transmission of the infection to date. However, bubonic plague (caused by *Yersinia pestis*) is being considered, as enlarged, bursting, and festering bubonic lymph nodes can be confused with the lymphogranular form of tularemia. It could also be bacillary dysentery (caused by *Shigella dysenteriae*); however, it must be admitted that we still do not have enough information to claim that the Hittites used biological weapons and, if so, which ones (18).

According to Homer, the spearheads and arrows of the warriors in the Trojan War (around 1200 BC) were dipped in biological poisons, but his information, like the entire Trojan War, cannot be reliably confirmed (19). Nor can the claims of some modern authors regarding the misuse of ergot alkaloids from *Claviceps purpurea* by the Assyrians during the expansion of their empire, which existed from 2025 BC to 609 BC, when the combined armies of the Medes and Babylonians conquered Assyria (20). Ergot alkaloids mainly affect the central nervous system and the vascular system and cause ergotism, which can be fatal. According to Das *et al*. (20), the Assyrians poisoned their enemies' wells with ergot. However, it is unclear how the Assyrians would have obtained ergot, as they did not grow rye, and how the wells would have been contaminated. During the so-called First Sacred War in Greece (595 BC - 585 BC), which concerned pilgrims' access to Delphi, members of the so-called Amphictyonic League, led by Athens, allegedly poisoned the wells supplying the port city of Kirra with water (21). They used the resinous plant black hellebore (*Helleborus niger*) for this purpose. Hellebore contains a number of alkaloids, glycosides, and steroidal saponins, which are very effective substances that cause incapacitation. A few hellebore seeds are enough to cause fatal poisoning, and the juice from the rhizome and above-ground parts of the plant causes burns to the skin and mucous membranes. When ingested, the poison causes stomach irritation, vomiting, and severe nausea. Whether hellebore can be used to poison water in wells is at least debatable.

Biological poisons and toxins seem to have been strategically effective in early historical times. According to Herodotus, during the 4th century BC Scythian archers dipped their arrowheads in decomposing human and animal corpses, most often snakes, or in blood mixed with manure (19). They relied on snake venom or the toxins of the pathogenic bacterium *Clostridium perfringens*, which causes gas gangrene, colic, watery diarrhea, or entero-toxemia, or the fairly common soil bacterium *Clostridium tetani*, whose toxin(s) cause tetanus, or lockjaw. In many parts of Africa, arrows with tips dipped in biological poisons were used in historical times. Various mixtures or extracts of plant or animal tissues were also used to kill, hypnotize, or incapacitate the enemy. The preparation of these mixtures was the prerogative of shamans or indigenous healers and combined shamanic, religious, and military practices. In many cases, this practice was effective, as the Portuguese discovered when they arrived in Senegal in 1444 and Sierra Leone in 1462 (22,23). John Hawkins (1532-1595), an English naval officer, pirate, and slave trader, also witnessed this when he landed with three ships in the Sierra Leone area in 1562 to capture people and trade them as slaves in the cities of Hispaniola in the Caribbean Sea (24). Biological poisons and toxins are, by today's standards, on the borderline between biological and chemical (toxin) agents, but as such they are classified as biological agents of biological origin.

Until the discovery of bacteria by the natural scientist Antoni van Leeuwenhoek (1632-1723) and the understanding of their association with disease in the mid-nineteenth century by Louis Pasteur (1822-1895), the purpose of ingesting biological material as BW was not always clear. During the Peloponnesian War (430 BC – 426 BC), when the city-state of Sparta was at war with the city-state of Athens, the ancient world was hit by the first real pandemic caused by a living microorganism, known as the Athenian plague. It first appeared as an epidemic

in southern Ethiopia and then spread as a pandemic to Egypt, Libya, Persia, and Greece, and was introduced to Athens via the port of Piraeus (25). It killed more than a third of the population, significantly weakening the Athenians and hastening their defeat by the Spartans. It was probably not a real plague. The Greek historian and politician Thucydides (460–455 BC – c. 399 BC) described a number of symptoms quite accurately, but these correspond to a whole range of infectious diseases. A number of later authors attempted to identify the disease: measles (John F. D. Shrewsbury (1898–1971) and Denys L. Page (1908–1978)), typhoid fever caused by one of the rickettsiae (William MacArthur (1884–1964)), typhus (Harold D. Evjen (1929–2016)), smallpox (Robert J. Littman), or Lassa fever (J. M. Hopper) (26–30). However, there are theories that bioterrorism was behind the outbreak of the Plague of Athens. This is stated by Manolis J Papagrigorakis in his 2013 study "The plague of Athens: an ancient act of bioterrorism?", where he refers to Thucydides' account of the poisoning of water reservoirs by the Spartans (25). The author also mentions the bacterium *Salmonella enterica* serovar Typhi, which was used as a biological weapon and was the cause of the plague in Athens (25). DNA analyses carried out to date have not provided an unambiguous answer, and the theory of military misuse of the bacterium has not been confirmed.

Various snakes and scorpions, and even bees, have been used as biological weapons. Prusias I (243-182 BC), the ruler of Bithynia (a region in Asia Minor adjacent to the southern coast of the Black Sea), waged war around 190 BC against neighboring Pergamon, ruled by Eumenes II Soter (221-158 BC). The Bithynian fleet was led by Hannibal of Carthage (247-183 BC), who at that time was an outlaw pursued by the Romans. Although the Pergamum ruler's fleet had numerical and technological superiority, Hannibal won thanks to the use of amphorae, which looked harmless but were filled with poisonous snakes and scorpions (31,32). The Pergamum fleet was then easy prey for Hannibal. Although the Roman emperor Septimius Severus (146-211 AD) was a capable military leader, he failed to conquer the Parthian fortress of Hatra (near Mosul, Iraq) in around 198 AD. The defenders threw clay pots filled with live scorpions at the Roman army. Roman Emperor Trajan (Marcus Ulpius Traianus, 53-117) fared similarly at this fortress, which he also failed to conquer. The use of bees directed at the enemy is mentioned by the Byzantine emperor Leo VI, known as the Wise or the Philosopher (866-912), in his theoretical military treatise Tactica (33).

Literary works contain countless examples of the use of human corpses, animal carcasses, various rotten waste, feces, and contaminated fabrics and objects for military purposes. There is also a wealth of information about the use of plant toxins, poisons, and other toxic substances to create biological weapons in ancient and medieval times. These were used in Asia, Europe, America, and Australia. However, much of this information cannot be verified today and thus remains in the realm of conjecture, conjecture, and speculation.

Literary sources state that during the Hundred Years' War between England and France, which lasted from 1337 to 1453, decomposing animal carcasses were used as biological weapons during the siege of the fortress of Thun-l'Évêque (1340) (34). In the 14th century, the plague returned to Europe after many years, earning the name Black Death. Between 1347 and 1353, an estimated 75–200 million people died of the plague. According to available information, this plague pandemic originated during the siege of the port of Caffa by the Tatar army of Jani Beg (35). During the siege, the plague broke out among the Tatar army, and the Tatars used the bodies of the dead as a biological weapon, catapulting them over the walls into the city. Genoese and merchants fleeing Crimea then carried the infection to Europe (36,37). Today, the fortress of Caffa lies on the outskirts of the city of Feodosia in Crimea. However, it is not certain whether the aim was to attack with biological weapons or simply an unorthodox attempt to get rid of the corpses. In 1422, during the siege of Karlštejn Castle in Bohemia, Hussite attackers catapulted dead bodies over the walls, but these were not infected with the plague. It is also reported (38) that they used the contents of two thousand carts loaded with manure, feces, and other waste for the catapults. A similar tactic of throwing plague victims into cities was allegedly used by the Russians during the siege of Reval, today's Tallinn, in 1710 (39–40). A 5,000-strong Russian army besieged the city on August 22 of that year; however, the plague epidemic had been in full force since August 10.

It is difficult to imagine that the Hussite fighters dragged two thousand carts just to reverse the course of an unsuccessful five-month siege of Karlštejn Castle. Similarly uncertain is the information regarding attempts to use biological elements as weapons in the following centuries (38–40). The claims are mostly based on data published many years after the event, and moreover, the primary information mostly comes from authors who were biased against the affected defenders. In many cases, the claims are based on local epidemics that were occurring at the time

or subsequently. It cannot be assumed that before the 20th century, the level of knowledge about the biology of microbes or their transmission from one host to another was sufficient to enable the targeted use of biological materials as BW. If further evidence is needed to support this argument, we recommend W. Seth Carus's comprehensive publication entitled "The History of Biological Weapons Use: What We Know and What We Don't" from 2015, which provides a detailed analysis of opinions on the validity of literary data concerning the misuse of biological agents since early history (41).

Actual problems with BW and efforts to ban them

During the nineteenth and first half of the twentieth century, advances in the understanding of the world of microorganisms and in the technical sciences progressed to such an extent that it became possible to realistically consider the targeted construction of biological weapons. According to some historical review studies, for example Britain, besides Germany, have experimented with or considered the use of biological agents during the World War I, but there is no evidence of systematic use comparable to German operations (by Battle Zone Editorial, Biological Warfare - Historical Examples of Biowarfare: Lessons from the Past. September 10, 2024). Moreover, the German operations were not primarily targeted at humans but at livestock, especially horses, which were the main driving force behind the logistical support of the fighting armies. The biological agents that dominated were the causative agent of anthrax (*Bacillus anthracis*) and the causative agent of glanders (*Burkholderia mallei*) (42). The question is whether the typhus epidemic, which had a significant impact on the conduct of the war, was caused intentionally. It is not clear whether the typhoid epidemics were caused by *Salmonella typhi* or *Rickettsia prowazekii*; and whether there were also diseases caused by *Bartonella quintana* or *Borrelia recurrentis* at the same time (43).

Based on the experience of World War I, the "International Convention for the Prohibition of the Use of Gases and Bacteriological Weapons in War," known as the Geneva Protocol, was signed in Geneva, Switzerland, on June 17, 1925. Thirty-eight countries signed this convention. According to this convention, it prohibited the first use of chemical or biological weapons in a state of war. However, it was not agreed that the ban would also apply to the production, storage, and transport of such weapons by the signatory countries (an extension of the convention in this sense was not signed until April 10, 1972, in Moscow, London, and Washington, D.C.; the new document was called the "Convention on the Prohibition of the Development, Production and Stockpiling of Bacteriological (Biological) and Toxin Weapons and on their Destruction"). The compliance of the signatory states with the Geneva Protocol is subject of controversy. France, as the depositary state, complied with the Geneva Protocol, but retained its scientific capabilities with a view to defending itself against an attack using biological weapons. However, given the situation in Germany, tests involving the spread of non-pathogenic bacteria had been carried out in the Paris metro since 1934, and a specialized laboratory for biological agents was opened in 1937. Research focused on defense against misuse or military use was significantly accelerated in 1938. This research resulted in the inclusion of certain toxins on the list of potentially misused biological substances and the verification of the possibility of their aerosol application (44). Great Britain, which ratified the Geneva Protocol in 1930, began, like France, to prepare for the possibility of war involving the use of biological weapons, given the situation in Europe. In 1936, the British Imperial Defense Committee established a Subcommittee on Bacteriological Warfare. As Canada was a British dominion, it coordinated issues with Great Britain related to biological agents, including carriers. Later, cooperation was extended to the United States. After the fall of France in 1940, experimental laboratories were set up at the Porton Down military site near Salisbury, where the Chemical Defense Experimental Station had existed since World War I, to study the use of anthrax (*Bacillus anthracis*) to kill animals in Germany and thus damage German agriculture. Work was also done on the possibility of aerosol application of anthrax. In the summer of 1942, an experiment with the aerosol application of biological agents was carried out on the island of Gruinard near the Scottish coast (45). The bomb was suspended about 1.5 meters above the ground and detonated electronically. In October 1942, another test was carried out on the deserted coast of Wales. A Blenheim bomber dropped a biological bomb from a height of 1.5 km, which landed 18 meters from the target point, from which groups of sheep were placed at a distance of approximately 110 and 300 meters. All the sheep died of anthrax (46). Both tests confirmed that warfare using biological weapons was possible and that BWs filled with anthrax was 100 to 1,000 times more effective than any chemical weapons available at the time.

Japan also worked on the production of biological weapons and their possible application in warfare. Dr. Ishii Shiro played a key role in Japan's program of research, large-scale production, and use of biological weapons,

declaring that "if BW weapons were placed on the list of prohibited weapons, then Japan should actually possess them and thus gain an advantage over its opponents in future wars." Japan had four offensive operational units focused on biological warfare. With auxiliary units, 15,000 people were available for the use of biological weapons in warfare. Of the four units of the Imperial Japanese Army, the best known is Unit 731, headquartered in Pingfang, one of the districts of Harbin, the largest city in the Japanese puppet state of Manchukuo (now northeastern China). The laboratory facilities of the unit were built between 1934 and 1939, and the original name was "Kwantung Army Epidemic Prevention and Water Purification Department," but it did not receive its numerical designation until 1941. During the Second Sino-Japanese War (1937–1945) and World War II, Unit 731 carried out horrific experiments on living humans, thus participating in the worst Japanese war crimes (41). However, it has been proven that only the Japanese army used BW during World War II.

In the post-war years, numerous accusations of BW use emerged, but most of them are unverified. One such accusation comes from Ken Alibek (*1950), who was one of the leading figures in Soviet BW research and who claimed that the Soviet Union used the intracellular bacterium *Francisella tularensis* to cause an epidemic of tularemia during the Battle of Stalingrad (autumn 1942 – winter 1943) (47). However, Alibek's claim has a number of opponents who propose an alternative explanation, namely that members of the Wehrmacht were infected by transmission from Russian civilians (48). However, this alternative is also not credible, as human-to-human transmission of tularemia is not known. Probably the most likely explanation is that tularemia was endemic in the Stalingrad area. Small rodents are natural hosts, which become a source of infection after they die. It is conceivable that during the fighting, explosions created aerosols containing infected particles from dead rodents, which became a source of inhalation infection. Nevertheless, the victorious powers did not end their experiments with highly dangerous microorganisms. This was essentially a consequence of the Cold War, when the United States, the United Kingdom, and the Soviet Union wanted to be prepared for the outbreak of a new "hot war." It is difficult to evaluate research in the field of biological agents during this period because information on individual research programs and any incidents that may have occurred is classified to varying degrees. Already during the Korean War (1950-1953), suspicions arose about the use of biological weapons, but documents from the Chinese and North Koreans were never made public (49,50). However, there is more information available about the biological programs carried out in the Soviet Union. These programs are summarized by Milton Leitenberg, Ken Alibek, and their colleagues (51,52,48). The United States military biological program was launched in 1942 with the official goal of developing defensive measures against a possible biological attack. The program was carried out in laboratories at Camp Detrick in Maryland, which was renamed Fort Detrick in 1956. Fort Detrick was the center of the US biological weapons program from 1943 to 1969. In November 1969, President Nixon announced the end of offensive research and production of biological weapons (53). The Fort Detrick base was transformed into the US Army Medical Research Institute of Infectious Diseases (USAMRIID), which still exists today. The British offensive biological program was terminated between 1955 and 1956 (54).

Given the threat of the Cold War turning into a hot war, the Convention on the Prohibition of the Development, Production and Stockpiling of Bacteriological (Biological) and Toxin Weapons and on their Destruction (55) was signed in London, Moscow, and Washington, D.C. It was to be one of the instruments for eliminating the use of biological agents as weapons of mass destruction. The instruments of ratification were deposited with the depositories of the Convention, the governments of the United Kingdom of Great Britain and Northern Ireland, the United States of America, and the Union of Soviet Socialist Republics, on April 30, 1973. The Convention entered into force on March 26, 1975, pursuant to Article XIV, paragraph 3. On that date, it also entered into force for the Czechoslovak Socialist Republic, as the Federal Assembly of the Czechoslovak Socialist Republic had approved the Convention and the President of the Republic had ratified it.

The Convention is valid indefinitely according to Article XIII, but it has been violated. Although representatives of the Soviet Union signed this "Convention" at the UN in 1972, anthrax spores leaked from laboratories belonging to the "Biopreparat" institution in Sverdlovsk in 1979. However, all documents relating to this incident were destroyed by the KGB (56). The laboratory technology and know-how relating to BW research that took place during the Cold War did not disappear. Biological research centers, such as the Vector laboratory in Koltsovo in southern Siberia, laboratories in Stepnogorsk and Obolensk (former USSR), USAMRIID laboratories in Bethesda (USA), Fort Detrick and Edgewood, both in Maryland (USA), the laboratories in Porton Down (UK) and FOI in Umea (Sweden), and the laboratories in Těchonín (former Czechoslovakia), have all focused-on biological

research on microbes which are in the list of biological agents that can be misused for military or terrorist purposes, however, always officially declared as defensive research. While the aforementioned laboratories in the United States, Sweden, the UK and the Czech Republic openly cooperate in the field of biological research, the same cannot be said about the laboratories in the former Soviet Union.

Research into biological agents that can be misused for military or terrorist purposes continues to this day. With the emergence of new laboratory technologies focused not only on molecular biology, BW research has been carried out in several directions, including (1) detection of the use of biological agents, (2) their identification and elimination from target structures, and (3) research focused on protecting people from BW, primarily research on filters, antibiotics, and vaccines. However, it has become apparent that microbiological, immunological, and infectious biological research of this kind is common throughout the civilian sphere and is essentially very difficult to classify as BW research, even in the case of microorganisms from the list of bio-agents of military importance. Many of these infections are zoonoses and occur naturally in certain areas of the world, and given the existence of developed tourism, it is relevant to address them even if such infections do not occur in the country where such research is conducted. Therefore, the whole issue of identifying the development, production, and stockpiling of BW has gradually shifted to addressing the question of whether the research being conducted is offensive in nature, i.e., aimed at developing BW, or defensive in nature, aimed at improving the protection of the population against infections, including efforts to eradicate the most dangerous ones. However, the problem arose of how to distinguish defensive research from offensive research, and what criteria to choose.

Advanced science and technology can influence the misuse of biological agents in several ways. It is important to talk about this only at the conceptual, policy, and risk assessment level. Easier access to powerful biotechnologies such as high throughput sequencing, automated DNA synthesis, and CRISPR have dramatically lowered the barrier to entry for biological research but it also raises concerns about dual use potential. Advanced techniques allow biological systems to be modified faster and more precisely than ever before. Synthetic biology enables the design of new biological constructs. AI assisted protein design and modelling can accelerate understanding of biological functions. However, the speed of innovation can outpace regulatory frameworks and oversight. Moreover, automation and "bio industrialization" reduce skill barriers needed for biological laboratories and as such increase the importance of strong governance and monitoring, and Digital biology (genomes, plasmid maps, and protocols online) enables global collaboration and raises concerns about misinterpretation or misuse of biological information. Given the general availability of advanced molecular biological techniques and bioinformatics databases, it cannot be ruled out that a certain state, certain group or individual will pursue creating and using biological warfare. In recent decades, bioterrorism has arisen, which is quite different from the concept of BW and biowarfare, but the means and tools are similar.

Final remarks

In an internal document (BWC/AD HOC GROUP/WP) from the Ad Hoc Group on Review of the Conclusions of the Biological and Toxin Weapons Convention (BTWC) originating from negotiations in 2001 and 2002, there is a proposal for six areas that would characterize potentially dangerous technology related to the development of BW. These are procedures aimed at: a) increasing the infectivity, pathogenicity, and antibiotic resistance of a given pathogen or increasing its resistance to the host's immune defense, b) improving the viability and maintaining the virulence of the pathogen during prolonged storage or release into the normal environment, c) facilitating the spread of biological agents in the form of fine particle aerosols, d) facilitating the dissemination of biological agents through contamination of food or water sources, e) creating new pathogens or modifying existing ones to evade current detection methods or the host's immune defenses, and finally f) assembling a chain of oligonucleotides for insertion into the genome of a pathogenic microorganism. At meetings of the VEREX, which was created to objectively evaluate measures for compliance with the Convention from a scientific and technical point of view, 21 potential measures were assessed during the proceedings that would distinguish between permitted (defensive) and prohibited (offensive) activities in biological research. However, none of the measures discussed would, on its own, be able to clearly distinguish between a defensive and an offensive biological program. A similar conclusion was presented at the European Defense Agency symposium on biological detection, identification, and monitoring in Munster, Germany, in June 2008. It was stated that no single technology is capable of reliably identifying biological agents in the event of their misuse (57). Roger Roffey, an expert at the FOI Swedish Defense Research

Agency, writes in the chapter "Biological weapons and potential indicators of offensive biological weapon activities," SIPRI Yearbook 2004: Armaments, Disarmament and International Security and lists a number of distinguishing points for the areas of "R&D, testing and evaluation," "agent production and storage," and finally "weapons, delivery systems, R&D, testing, filling, and storage" (58). However, Roffey concludes by noting that only complete transparency in the rapidly developing field of biological defense research and development is essential, particularly for building trust among states that new technologies are not being misused. Milton Leitenberg (a US expert on the misuse of biological agents from the Center for International and Security Studies at Maryland) comes to a similar conclusion in his article "Distinguishing Offensive from Defensive Biological Weapons Research" (59). Distinguishing between offensive and defensive research at all levels of research and development is very difficult, as evidenced by a number of misjudged research and intelligence reports submitted to political representatives. It can therefore be clearly concluded that only transparency in biological defense research and development programs is essential for building trust between states. In addition to transparency, there must also be the possibility of control and verification using reliable technical means, enabling the implementation, if necessary, of the Bio DIM concept (biological detection, identification and monitoring) introduced, for example, by the European Defence Agency.

There are not many options protection from possible hidden actors using dual-use biotechnology. In the fight against these risks, it is possible to focus efforts on biosecurity (prevention of misuse), management and control of dual-use research, strict application of the ethical framework for synthetic biology, or control of the export of sensitive biological research items. The aim of all these approaches is to ensure that scientific progress remains safe, beneficial and globally responsible. From a global perspective, strengthening the position of the BWC is key. The BWC has legal force as an international treaty, but it does not have a formal verification regime, as states have never agreed to one. This means: the commitments are legally binding, but their control is politically and technically weaker than in other disarmament treaties. In the case of the BWC, it relies on so-called confidence-building measures (CBMs) - voluntary reporting of activities that are intended to increase transparency. As a result, the BWC is often described as “normatively strong but institutionally weak”. The Ninth Review Conference of the BWC established a working group to strengthen the convention, which is to reopen the issue of verification (see Final document of the 9th Review Conference, Geneva, 28 November-16 December 2022). However, there is still no consensus - in particular, the USA, Russia and some other states have different ideas about what verification should look like.

In conclusion, we can only state that the literature on the misuse of biological agents, including documents related to the Convention, demonstrates the impossibility of a general consensus on the non-misuse of microorganisms and their toxic products for nefarious purposes. In the text, we stated that even during the negotiations of state representatives regarding the amendments and clarifications of the BWC, consensus was not always reached. All this proves that the issues of biological security are very complex and in the current world with dynamically ongoing conflicts, the issue of biological security needs to be further investigated and the importance of the BWC reminded. Current knowledge does not provide a clear guarantee of biological security.

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AM - writing original draft, review and editing;
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