DEVELOPMENT OF KNOWLEDGE AND SKILLS FOR BOTH TEACHERS AND STUDENTS THROUGH EXTRACURRICULAR EDUCATION MATERIALS ON BIOSAFETY AND BIOSECURITY

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Abstract

Today's learning strategies based on life skills, transversal and professional competences, are some of the modern forms of education, which can flexibly respond to the historical moment faced by the education system in the time of biotechnological revolution. The immense achievements in the field of biotechnology have brought the need for educators to develop competence based extracurricular materials on the field. Developing the future competences of the student's in this field means not only addressing the positive impact of biotechnology in human progress, but also raising awareness on the risks of its use for harmful purposes, against the humanity.

Biosafety and biosecurity extracurricular modules can be used and delivered not only to students in High Schools but also to multiple audiences with a very specific goal. Education materials in this field serve to ingrain relevant principle for guiding effective decisions and behavior; to present and highlight the importance of biosafety and biosecurity, and to recognize the impact of their use for a secure future of the planet and life on it.

Variations in biosafety training requirements, incident-reporting practices, and attitudes toward laboratory safety…support the development of core competencies in biosafety practices that could lead to more uniform and robust safety culture.

In this material we are proposing together with the learning objectives the content that provides the best learning experience for both student and teachers, the timing for each activity, materials that can be used like background documents, and a "common language" between the content and the teacher that will attract the attention of the users. This extracurricular biosafety material offers great promise in the development of autonomous and enduring biosafety knowledge and skills that are relevant, useful, and resilient for both teachers and students.

Keywords: *biosafety, biosecurity, biological war, extracurricular module*

1. INTRODUCTION

The well-known scientist Stephen Hawking had a pessimistic point of view on the future of human life on Earth, quote "... I don't think we are going to survive more than 1000 years without leaving this fragile planet..." According to him, traveling in space for finding a new home in another planet was a necessity due to the risk of nuclear war and genetically modified organisms. "... The last (GMOs) are the most dangerous because we know little things about them" - Hawking says. Genetically modified organisms, and not only them can lead to a very dangerous war, biological war [1].

Developments and achievements in the field of biology in general and that of biotechnology in particular have brought not only innovation and progress, but also a number of problems of an ethical and existential nature. Based on advances in biotechnology today there are a number of organisms that can be exploited by humans for a range of activities, but if the process it is not properly handled their use can cause harm, either directly, or indirectly, resulting in considerable health, environmental, social, and economic losses. As a result, there is global recognition for the need to develop international biosafety and biosecurity capacity not only for professionals but for everyone and education system is the perfect place for this development of knowledge and skills [2].

Various international agreements such as the International Health Regulations, 2005; United Nations Security Council Resolution 1540; the Biological and Toxin Weapons Convention, 1975; the

International Food Standards of the Codex Alimentarius Commission 1999 (FAO, 1999); and the Cartagena Protocol on Biosafety to the Convention on Biological Diversity, 2000 (CBD, 2000), require the provision of relevant education and training, are put in place to prevent harm from biological material.

Crucial to the implementation of such extracurricular materials is an approach in which the modules can be released as a stand-alone program for the students or used as accredited components in post-graduate degree programs for teacher formation. An integrated biosafety and biosecurity curriculum would therefore enable students to build their capabilities to effectively address the bio risk threats. In order to ensure that all of the key biosafety and biosecurity concepts are covered, it is necessary that such an extracurricular module is developed as part of a broad-based, cohesive, and comprehensive core curriculum, encompassing all elements common to biosafety and biosecurity regulation. The design of such module encompasses not only with the need of the school to build competence based learning, but also with the curriculum of biology and natural sciences that are related to contemporary concepts like biosecurity, public health, natural resource management, bio control, and bioremediation. In fact, the breadth of coverage offered by this module was of especial focus, so that it appeals to students with interests and motivations. The most obvious benefits from utilizing this module in biosafety and biosecurity include flexible approach**,** comprehensive coverage, user-friendly by taking into consideration user's individual learning styles at minimal expense, and staff time investment.

2. METHODOLOGY

The methodology of this paper is designed to impart to students of Secondary Education basic knowledge about biosecurity and biosafety. The module consists of four activities, which give teachers the opportunity to develop interesting lessons or as topics for the development of portfolio tasks. Instructions for the teacher's and student's tasks are given in each activity. The design of the activities of this module is based on the information collected and analyzed from scientific articles related to the topic, as well as from data provided by the publications of WHO, which are adapted for high school students with a simple and comprehensible scientific language.

To build the curriculum we have considered the following steps:

Identification of knowledge, skills, habits, tasks and information that a subject must undertake during the school cycle; 2. Grouping according to their specificity of knowledge, skills, habits, tasks and information, classifying them into logical categories; 3. Defining programs and their basic topics; 4. Determining the expertise, skills and knowledge that teachers should have that will transmit this curriculum, seeing these, not only in the scientific and pedagogical direction, but also as communication skills, organizational skills and being responsible; 5. Communication of the curriculum and the programs that support it, their implementers; 6. Establishing and maintaining an open and ongoing dialogue with curriculum implementers with the aim of continuously improving it [3].

Based on the theoretical material, the worksheets for the development of the lesson were prepared. Each activity contains ideas, exercises and concrete examples of the tasks that the student must fulfill, as well as the role that the teacher has in each of them. They come with illustrations, figures and posters to make the reader as clear and attractive as possible, as well as easier to implement.

3. THE HISTORY OF BIOLOGICAL WAR

Biological war has started earlier than we think. The history is filled with examples of biological agents used for war purposes. During ancient times poisoned arrows going through decomposed bodies were used as biological weapons. On Greek, Persian and Roman literature are given examples of dead bodies used with intention to contaminate drinkable water sources [4]. During Indian war in 1763, British forces distributed contaminated blankets, which had been used from smallpox victims, in order to spread this disease inside Indian population [4]. In World War I, German army developed anthrax, cholera and a wheat fungus in order to be used as biological weapons. They spread the plague in Saint Petersburg

using infected mules [4]. The World War II, Japanese forces operated a secret biological research facility in Manchurian. They exposed more than 3000 prisoners to plague, anthrax, syphilis and other potential agents, in order to observe and study those diseases [4]. During the Cold War, both the Soviet Union and the United States developed programs of biological weapons. These programs were legally stopped in 1972 with the signature of Biological Weapon Convention. In 1991, Russian journalist Boris Yeltsin confirmed that Soviet Union had broken the Convention because it had developed a project of clandestine biological weapons [4]. In 1984 a bioterrorist attack happened in Dallas, Oregon. The salads at ten local restaurants were contaminated with *Salmonella typhimurium.* The author was Bhagwan Shree Rajnesh, the head of an Indian spiritualist cult. A total of 751 people was affected, but no fatal damage was registered [5]. In 2001 anthrax was delivered by mail in governmental and media offices of the US. 22 people were affected and five of them died as a result. In December 2002, six terrorists and one chemist were arrested in Manchester, UK, their apartment was serving as a "ricin laboratory". [5]. In February 2004, three U.S. Senate office buildings were closed after finding the toxin ricin into the mailroom serving Senate Majority Leader Bill Frist's office [5].

4. BIOLOGICAL WEAPONS AND BIO WARFARE

The Center for Disease, Control and Prevention (CDC) defines bioterrorism as the act of "deliberate spread of viruses, bacteria or other reproductive organisms which can cause diseases or be fatal for humans, animals and plants" [6].Biological weapon is considered every living organism or substance derived from living organisms, which can be able to cause disease on human beings, plants or animals [1] .There exist 5 different types of biological agents that could be used as weapons in biological war or terrorist attacks. These include [1] :

- Bacteria – single-cell organisms able to cause different diseases, such as anthrax, plague, brucellosis, tularemia etc.
- Viruses – intracellular parasites that can cause diseases such as Venezuelan equine encephalitis.
- Rickettsia- microorganisms that resemble bacteria but differ in that they are intracellular parasites that reproduce inside cells. Typhus and Q fever are examples of diseases that rickettsia can cause.
- Fungi – pathogens that can infect crops to cause such diseases as rice blast, cereal rust, wheat smut and potato blight.
- Toxins – poisons that can be used after extraction from snakes, insects, spiders, marine organisms, plants, fungi, bacteria and animals.

The most typical ways of contamination are [6]:

- Through breathing–all laboratory processes that produce aerosols (spinning, mixtures, decantation of fluids etc.)
- Through direct contact–contaminated needles or sharp objects able to cause skin bloodshed
- Through the mouth–eating or drinking into a contaminated space, contact of contaminated hands with the mouth.

Biological weapons have some advantages compared with other types of weapons. They are fairly cheap to produce, don't need a sophisticated technology or considerable space and can be easily transportable. Also, they can be "detonated" and due to the long time, it takes for them to spread and make effect, there is plenty of time for the perpetrator to escape undetected. Biological weapons can be difficult to control or predict in a battlefield situation and they have the potential to cause great harm [6].

The US Centers for Disease Control and Prevention (CDC) has made also a classification of bacteria and viruses (Table 1.1) in order to study and understand their risk and impact on public health [7].

Tab 1.1 Classification of biological agents based on their risk rate

	Category A	**Category B**	**Category C**
Criteria	• High mortality • Easily distributed in the population • Requires special intervention	• Average distribution and morbidity	• Easily produced under lab conditions • Potential for morbidity and mortality
Example	• *Bacillus anthracis* • *Clostridiumbotulinum* • *Francisellatularensis* • *Variolamajor* • *Yersiniapestis* • Ebola virus • Marburg virus	• *Brucella sp.* • Ricin toxin • *Coxiellaburnetii* • *Salmonelasp.* • *Vibriocholerae*	• Yellow fever virus • Hantavirus • *Mycobacteriumtuberculosis*

4.1 Bacillusanthracis (Anthrax)

Bacillus anthracis bacterium is one of the most fatal agents to be used as a biological weapon. The gram-positive, rod-shaped anthrax spores that are found naturally in soil, can be produced in a lab and lasts for a long time in the environment. Anthrax has been used as a bio-weapon for almost a century mixed with powders, sprays, food and water. The invisible, infectious, odorless and tasteless spores of *Bacillus anthracis* make it a flexible bio-weapon [8].

4.2 Clostridium botulinum toxin (Botulism)

Botulism is a serious muscle-paralyzing disease caused by a nerve toxin produced by a bacterium called *Clostridium botulinum*. The bacteria is found naturally in forest soils, lakes and streams deep sediments as well as in the intestines of certain fishes and animals [8]. All four forms of botulisms (food borne, infant, wound and animal) induce illness through a common pathway. They cause muscle weakness, difficulty in speaking and swallowing and double, blurred vision. Botulinum is relatively easy to produce and has extreme power and lethality. It can be distributed via air or by contamination of water and food supplies. A gram of Botulinum toxin can kill more than a million people if inhaled [8].

4.3 Variola major (Smallpox)

Variola major virus causes smallpox, a highly contagious and infectious disease that has no cure and can be only prevented by vaccination. The virus can be transmitted directly from person to person or indirectly from an infected person, via contaminated items. A deliberate release of smallpox is a great threat. The Soviet government started a program in 1980 to develop smallpox virus in large quantities stored in refrigerated tanks in order to use it as a bio-weapon. The threat of smallpox being used as a biological weapon decreased when WHO launched a successful global immunization program against Smallpox in 1967 [8].

4.4 Francisella tularensis (Tularemia)

The extreme infectiousness, the rate of dispersion and ability to cause illness and death make *Francisella tularensis* bacterium a dangerous bioweapon. People affected with *Francisella tularensis* experience symptoms such as skin ulcer, fever, cough, vomiting and diarrhea [8].

4.5 Yersinia pestis (Plague)

Yersinia pestis bacterium causes pneumonic plague and can be created in a laboratory in high quantity. Pneumonic plague spreads from person to person and causes symptoms such as fever, weakness and pneumonia in early stage and, if not treated early, leads to respiratory failure, shock and death [8].

4.6 Ebola virus

Ebola virus disease (EVD) is a deadly disease induced by infection with one of the Ebola virus forms. Ebola was first discovered in 1976 in the Democratic Republic of Congo and is transmitted to humans from wild animals, causing an average case fatality rate of 50-90%. This virus can be transmitted by direct contact with blood, biological fluids of the body and infected tissues of humans and animals. Symptoms include: fever, headache, muscle and joint pain, weakness, diarrhea, vomiting, stomach ache and lack of appetite. There is currently no vaccine licensed for this virus, but many laboratories are conducting research and testing some types of vaccines. The Russian Federal Biomedical Agency has expressed concerns that the Ebola virus could be used as a biological weapon. The same concern is shared also by Dr. Peter Walsh from University of Cambridge who says that "... an extremely large number of deaths can occur if a certain group decides to create a destructive bomb with the virus and use it in a highly populated public area. As a biological weapon, Ebola virus poses a great threat to human life because of its high case-fatality rate [8].

4.7 Marburg virus

The *Marburg Virus* causes the Marburg Hemorrhagic Fever. The Virus belongs to the filo virus family just like the Ebola virus. It was first recognized in 1967. The first cases of hemorrhagic fever were detected simultaneously in Marburg, Frankfurt (Germany) and Belgrade (Serbia). People infected had been exposed to imported African green monkeys or their tissues while conducting researches in the lab. The Soviet Union conducted experimentations with the Marburg virus form to transform it into a biological weapon [7].

5. ACCESS TO "OPEN DATA" INFORMATION AND TECHNOLOGICAL DEVELOPMENTS

It is estimated that hundreds of "open" scientific articles and research studies are published online each year at International level. That easy access to information certainly helps the development of science and technology, but also increases the risk of using that knowledge for developing biological weapons by people far beyond the scientific community [9].

In the fall of 2001, Dr. Eckard Wimmer of the State University of New York, surprised the scientific community when he announced that his lab obtained synthetic poliovirus. The construction of the poliovirus in laboratory conditions consisted of collecting all copies of the 7741 bases that made up the virus. The process was time consuming, but straight-forward. In the moment that they had assembled the full-length cDNA, they transcribed it with RNA polymerase in order to make the single stranded RNA genome of poliovirus. After that, they made live virus by incubating the RNA in an extract from HeLa cells. When injected into mice, the animals developed a neurological disease both chemically and histologically indistinguishable from poliomyelitis. In the end, the authors concluded that they had indeed created a synthetic copy of poliovirus from scratch. This led to serious debate about the potential use of DNA synthesis technology in the creation of biological agents[10].

In 1997, Dr.David Edwards published a paper in "Science" where he described a revolutionary method to deliver medicines through inhaling, using large porous carrier particles. The method dramatically increased the amount of the drug that went deep into the lungs. His work consisted in the creation of several drug or hormone-dispersed carrier particles for inhaling. There is no doubt that this porous particle technology holds a huge promise for improving drug inhalers. However, this technological advance also presents a classic dual-use dilemma. Such inhalers can enable the transmission of anthrax, tularemia, Q fever, smallpox, viral encephalitis, viral hemorrhagic fevers or botulism [11]. In October 2005 a group of scientists, including Dr. Terrence Tumpey from the Center for Disease Control and

Prevention (CDC), Dr. Jeffery Taubenberger and Dr. Christopher Basler, published a paper in "Science". The paper described the recreation of the 1918 Spanish influenza virus. But, how did they manage to accomplish that [12].

Lung tissue samples, containing the extinct virus, were taken from archived autopsy materials and an influenza victim who had been preserved miraculously in Alaska. Scientists found that the recreated virus was very similar to influenza viruses that only infected birds. The now extinct virus had developed a mutation that made it possible to spread between humans. The complete sequences of the coding for all eight viral gene segments of the 1918 influenza virus are now part of the GenBank database. The publication of such experimental works is a concern about biological safety [12].

Various organizations or countries interested in developing biological weapons can cultivate antibiotic-resistant pathogens, which, if distributed to the population, can cause catastrophic damage. This is a major concern as long as the population and medical structures are unprepared to fight diseases or epidemics caused by genetically modified organisms [13].

Today, developments in the field of genetics and biotechnology have radically changed the way we conduct research. They have also increased concerns about integrating new technologies into biological weapons programs [9].

A gene-processing technology called CRISPR-Cas9 enables genome modification by manipulating DNA sequences. Used in safe hands, this technology allows the correction of defects and the treatment of genetic diseases; if misused, it is a source of big risks. Through CRISPR-Cas9 it is possible to control genes to be expressed in plants, animals or humans, delete unwanted features and add the desired features with a higher quality than the technologies used earlier [14].

This technique can be considered as a revolution, because it costs almost nothing and is very simple to use. In the past, thousands of dollars and months of work were needed in order to modify a gene. Today the same job takes few hours and is it costs only 75$ [14].

Moreover, the Cas protein acts on every possible tested organism. Its applications have been numerous, such as correcting genetic defects, treating and stopping the spread of diseases and improving food production. The concern that technology could be used to develop biological weapons is growing day by day [14]. In 2016, Bill Gates stated: "the next epidemic may be originated from the computer of a terrorist, who could deliberately use genetic engineering to create a synthetic version of the smallpox virus". In 2017, John Sotos of "Life Sciences and Health" stated that "...genetic modification-based research can open the doors for the development of biological weapons with unimaginable destructive potential". However, for the moment, CRISPR's threats are relatively modest, because working on the creation of biological weapons using CRISPR technology requires specific capabilities and facilities [15].

This does not mean that we should completely ignore the possibility of using CRISPR from unlicensed individuals. The potential of technology is to be explored by biologists and engineers and may inadvertently open the door to ethical issues in the future [15].

In February 2016, the Director of National Intelligence of the United States, James Clapper, published a report mentioning that: "the developments in genome modification during 2015 have enabled high-level biologists to intervene in the human cell line (cells responsible for reproduction), which can pave the way for heritable genetic changes". Considering all the advances in the field of genetics, as well as the developments of CRISPR technology, the potential for irreparable damage is daunting. As long as such changes in cells can be transmitted to subsequent generations important questions are raised, such as: **Do we have the right to make substantial changes that affect the future of generations without their approval?** Modifying the genomes of organisms and releasing them into the population without keeping them under control is like firing a weapon, the action of which cannot be reversed. Once a genetically modified organism is introduced and reproduced, the modified genes consequently become part of the population genome forever [6].

We can predict the effects of these genes on the appearance of phenotypes in the next 2 or 3 generations, but then it becomes difficult to predict and control what may occur in the future. This is because as time passes, the evolution of genomes becomes more complex [16].

6. BIOSAFETY AND BIOSECURITY

According to WHO (World Health Organization, 2006) Biosafety includes "the measures taken during the use of biological hazardous materials, for scientific research, with the aim of avoiding human or environmental contamination".

This is achieved through:

- Administrative controls
- Engineering controls
- Personal protective equipment (PPE)
- Special practices and procedures

In 2006, WHO incorporated a new concept that is different from the concept of "Biosafety", this concept is "Biosecurity". According WHO, "Biosecurity is about protecting the biological hazardous materials and sensitive information from theft and misuse". The concept of "Biosecurity" includes also *the physical security of personnel* (access only to authorized individuals, retina control, access cards, scanning and visitor control), the *security of transport and materials* (materials transferred between laboratories required information and control of materials, receiver recognition, physical security of the shipment and scanning of the individual who handles the transport), *information security* (safeguarding sensitive information from public spreading, controlling the network where information is exchanged, as well as communication security).

Biosafety and Biosecurity complement each other. While **Biosafety** provides a safe laboratory space assuring protection from the dangerous infections/diseases that can be caused by manipulating biological materials, **Biosecurity** ensures that any potential infection won't cross the barriers of the laboratory. The environment pollution and the misuse of scientific research are serious concerns as it is the potential risk of contamination by working in a research laboratory. This is why there is a need to create, improve and implement specific protocols and regulations that will avoid those risks. As long as there is no technology or process that can stop the use of biological weapons, the implementation of international non-proliferation agreements and partnerships remains a critical factor in achieving global health security [17].

Laboratories play an important role in biological safety and protection. According to WHO (2006) a bio safe laboratory is a laboratory that implements a set of technologies, practices and principles to protect against exposure or spread of toxins & pathogens. It includes labor safety practices and tools, the strict follow up of written working procedures and instructions, and the application of microbiological techniques.

Safe and secure laboratories [18]):

- Preserve substances that can spread infections
- Build public confidence in research activities undertaken by the scientific community
- Increase investor interest and transparency in the biomedical and biotechnological industry
- Preserve valuable scientific research that serves as a commercial asset
- Reduce the risk of bioterrorism

In the research laboratories, potentially dangerous biological agents that can cause human disease (viruses, bacteria, fungi, parasites or toxins derived from them) are manipulated. Such laboratories

should have special structures and extremely safe working practices in order to protect the staff, the community and the environment from any potential harm [18].

Poor and unsafe lab working practices can be the cause of various contaminations or diseases. This is the reason why biological laboratories can be a threat to the international community and the global public health when if they lack security conditions, material management (storage use, transfer and treatment of waste), staff training and qualification. Each biological laboratory should reduce biological risk at maximum. It should strongly consider all the consequences of every biological material considered hazardous going outside the lab, accidentally or intentionally [17].

There are two strategies of protection [18]:

- Creating Primary barriers: keeping the potential risky agent at source (i.e. animal cages, specialized equipment such as centrifuges or fermenters)
- Using Secondary barriers if the primary barrier is overpassed: store the agent inside the laboratory (a specific laboratory type, directed ventilation system, waste filtration, solid and liquid waste treatment).

According to the Center for Disease Control and Protection (CDC) and the United States National Institute of Defense (NIH) there are 4 levels of lab safety required in function of the danger of biological agents manipulated (Tab 1.2; Tab 1.3; Tab 1.4; Tab 1.5).

Tab 1.2 Characteristics of BSL-1- Biosafety Level 1Lab [19]

Lab description	Examples of microorganisms used	Lab practices and procedures
Biosafety Level 1: does not cause human disease, it requires basic techniques to work on. The lab may be part of another building, this type of lab is found in schools and colleges	Harmless microorganisms: *Escherichiacoli*, *Bacillussubtilis*, *Saccharomycescerevisiae* etc.	Basic techniques of protection include: hand washing, standard personal protective equipment (gloves, lab coat or gown and eye protection), biohazard signs, decontaminating surfaces and equipment at least daily, safe handling of sharps, prohibiting mouth pipetting, prohibiting food, drink and smoking materials in the lab

Tab 1.3 Characteristics of BSL-2- Biosafety Level 2 Lab [19]

Lab description	Examples of microorganisms used	Lab practices and procedures
Biosafety Level 2: moderate hazard to personnel and the environment, limited access	Moderate risk agents for human and environmental damage: hepatitis A, B and C viruses, HIV, pathogenic *Escherichiacoli*, *Staphylococcusaureus*, *Salmonella*, *Plasmodiumfalciparum* and *Taxoplasmagondii*	Personnel trained to work with pathogenic agents, laboratory procedures are performed in secure cabinets, eyewash station, lockable doors and more controlled facility access, additional personal physical protection equipment

Tab 1.4 Characteristics of BSL-3- Biosafety Level 3 Lab [19]

Lab description	Examples of microorganisms used	Lab practices and procedures
Biosafety Level 3: used for diagnostic purposes and research. Lab facilities must be separated from other buildings and should be equipped with two sets of consecutive doors (to minimize the risk of air leakage), ventilation system (clean air goes in areas where it is worked with pathogenic agents and then in the filtration system before recirculating).	Airborne microorganisms that can cause serious illnesses: *Francisella tularensis*, *Mycobacterium tuberculosis*, *Chlamydia psittaci*, Venezuelan equine encephalitis virus, *Coxiella burnetii*, *Rickettsia rickettsii*, yellow fever virus, West Nile virus, etc.	Lab personnel must be medically supervised and immunized (antibody equipment), working procedures are performed in secure cabinets, personnel must use specific protective clothing (aprons attached to the back), specific clothing should be used inside the lab and should be cleaned after each use, access is restricted and controlled all the time

Tab 1.5 Characteristics of BSL-4- Biosafety Level 4 Lab [19]

Lab description	Examples of microorganisms used	Lab practices and procedures
Biosafety Level 4: the highest level of precaution. This type of lab should be in a dedicated building or a completely isolated facility with dedicated air system vaccum lines and decontamination systems. Air exhaust and used water must pass through HEPA filtration before leaving the facility.	Easily transmitted biological pathogenic agents that can cause fatal diseases for which there is no treatment or vaccination: viruses causing hemorrhagic fever (Marburg, Ebola, Lassa), Hendra virus, Nipah virus etc.	The use of specific suits that cover the whole body and have positive air pressure, high safety cabinets (Class III). Lab materials coming out of cabinets are sterilized in autoclaves or with powerful disinfectants, staff always leave the lab after they have completed a laundry with chemicals for cleaning, removing the pressure suit in another room and taking a personal shower, all entrances and exits

What are the protection means in case of a biological attack? Clothes: most of the biological agents in the air cannot penetrate unless there are cracks on the skin. After an aerosol attack, taking off the clothes removes the largest surface of contamination. Then, washing skin with plenty of water and soap eliminates 99.99% of those microorganisms that may still be left on the skin [5]. Medical protection: the use of latex gloves, masks and goggles, aprons or protective clothing. The affected people should be isolated in separate rooms during treatment [5]. Masks: Recently, special gas masks or high-efficiency air-defense (HEPA) masks have been developed and used when exposed to tuberculosis [5]. Antibiotics: are used in order to fight the bacterium or virus [5]. Vaccination: is carried out to prevent infection of various diseases caused by biological agents, for example: anthrax, Q fever, yellow fever and smallpox [5].

7. DEVELOPING CURRICULUM MATERIALS AND GUIDES

Activity 1. Curriculum project

The purpose of the curriculum projects is to put in practice the theoretical information skills and knowledge that students have acquired during classes. This activity, leads students to a higher level of learning [3].

Topic: "Scientific laboratory and biological protection"

Project purpose: Practical application of the extracurricular module "Security and safety in the context of biological warfare" by fostering team spirit and increasing the level of learning and critical thinking.

Learning outcomes: At the end of this project students will be able to:

1. Practice the basic elements needed to have a safe laboratory
2. Specifically build a safe biological laboratory model
3. Apply a general safety regulation while working in the laboratory

Steps to follow:

Step 1: The teacher explains to the students the methodology on how to build a safe laboratory

Step 2: Students are divided into small groups of 4 and each group chooses to build a specific safe biological laboratory corresponding to a particular task (for example medicine production, working with stem cells, modifying bacteria etc.)

Step 3: Each member of the group plays a role as follows:

Member 1 provides the information for the implementation of the project (what is a safe laboratory, what are the safety conditions, the role of these laboratories in industry, environment etc.). He/she monitors the work progress of other members and writes a work report. The report contains: timeline of implementation, names of project participants and their tasks, project theme, objectives, process, outcomes and outputs. Information resources:

- WHO (2006), "Bio risk management-Laboratory biosecurity guidance"
- Osman Sh.A. (2004) "*Laboratory Biosafety and Biosecurity-Biohazard Danger"* http://www.apbtn.org/apbtn/trainers.html

Member 2 is responsible for drafting the regulation of laboratory work safety (with regards to clothes, behavior in the laboratory environment, treatment of biological samples, and procedure in the event of accidents during laboratory work)

Member 3 and 4 are responsible for designing a safe biological research lab. They can build a 3D model of this lab using available materials (like: wood, plastic, foam, playdough, etc.). They present their ideas on a laboratory environment where the focus is not on identifying work equipment, but the safety equipment. The outline or model shall indicate the locations of the first aid kit, fire extinguishers, exits and emergency stairs, alarm system, access control systems- exits, protective filters for the propagation of biological hazardous elements, biological data protection systems and equipment's etc.References that could be used:

NCBI (2011), "*Laboratory Security- Prudent Practices in the Laboratory"* https://www.ncbi.nlm.nih.gov/books/NBK55881

Stony Brook University, "*Laboratory Security- Environmental Health and Safety"* https://www.ehs.stonybrook.edu/programs/laboratory-safety/laboratory-security

Teacher's role: Monitors and controls step by step the performance of students. He/she guides, advices and support. The teacher invites other teachers, parents and students from other classes to observe and evaluate the project.

Evaluation:

Table 1.6 Qualitative and quantitative criteria on which the project evaluation is based

Evaluation	Criteria
-	The report is not submitted. The report has many errors, there no teamwork and deadlines are not respected.
±	Very few sources of information are used, students try to collaborate, deadlines are generally respected, linguistic errors and unstructured report
++	Different sources of information used, good use of knowledge, deadlines are respected, there is student cooperation, the report is linguistic error-free and well structured, there is lab posting through a poster.
+++	The student has used a wide variety of information sources (beyond those presented in the text), affirms independent critical judgments, report submission is correct and well structured, the regulation is rich and functional in accordance with the type of biological lab, lab model responds to the theoretical and practical requirement of a safe biological laboratory

Activity 2. The competition

Studies show that the competition encourages students to work harder, study harder and build confidence. Group competitions encourage team spirit and collaboration meanwhile individual ones reinforces the personal habits and skills. In the academic world, competition serves as a building block that transforms potential into success. During the competitions students experience the taste of victory and loss in life [20].

On a large table are placed 20 envelopes with questions on the topics "Biosafety and Biosecurity". Students are divided into two groups. Each member has the right to randomly take an envelope and respond within one minute. For each correct answer the group earns 3 points. If the student does not know the answer his group loses 2 points and the right to reply is passed to another group member. In case the student gives the wrong answer the other group is added 2 points and the opponents are given the choice. The groups draw the first lottery and each member has the right to choose. The order of response within the group is determined alphabetically.

Teacher's role: The teacher plays the role of the referee, who measures time and evaluates the answers given as correct or incorrect. He/she is impartial in judgment and does not favor a particular group.

Evaluation: At the end of the competition the teacher rewards the winning group (puts each member group extra points) and congratulates the other group on giving the courage to not give up, because the loss is part of the race.

Below are listed 20 questions that a biology teacher can use to organize an interesting competition. Questions and answers are part of the theoretical material.

1. What is bioterrorism or biological warfare?

The Center for Disease Control and Prevention (CDC) defines bioterrorism as "the deliberate spread of viruses, bacteria or other reproductive organisms that can cause disease or kill humans, plants or animals. This can be achieved: through air (aerosol sprays), water, food absorption or skin injection.

2. What does "Biosafety" mean?

Biosafety means the entire set of measures taken when using biological hazardous materials for scientific research, in order to avoid harming ourselves, others or the environment.

3. What does "Biosecurity" mean?

Biosecurity includes all the necessary measures taken to safeguard biological hazardous materials, as well as the protection of sensitive/discrete information from theft and misuse.

4. Is there any connection between "Biosafety" and "Biosecurity"?

There is a close link between Biosafety and Biosecurity. Both of these concepts complement each other; thus, if Biosafety provides a safe laboratory environment to protect against dangerous infections/diseases, Biosecurity ensures that potential infections do not cross the lab barriers into society and environment.

5. What do we mean by biological weapons and how are they divided?

Biological weapon is considered any kind of biological agent capable to cause disease to humans, animals or plants. There are five different categories of biological agents that can be used as weapons for terrorism purposes. These include: bacteria, rickets, viruses, fungi and toxins.

6. When did the use of biological weapons first begin and what are the most popular wars in which these weapons have been used?

According to the information provided by historians and researchers, infected arrows and dead bodies infecting water sources have been used in antiquity as biological weapons. The main wars were bioweapons have been used are: the war between France and India (1710), World War I (1914-1918), World War II (1939-1945) and the Cold War.

7. What are the advantages of using biological weapons?

These weapons require little capital to produce; even a poor state can develop biological warfare programs if they have a group of biologists and secret buildings where to accomplish this activity. Distribution also has facilities, because such weapons are difficult to be detected, do not occupy significant space and can be easily moved anywhere. They do not require any advanced technology to be created and can be applied in the battlefield by a single person.

8. List the most dangerous biological weapons.

The most dangerous biological weapons are: *Bacillus anthracis* (anthrax), *Clostridium botulinium* toxin (botulism), Variola major virus (smallpox), *Francisella tularensis* (tularemia), *Yersinia pestis* (plague), Ebola virus (Ebola Hemorrhagic fever) and Marburg virus (Marburg hemorrhagic fever).

9. How are biological agents classified according to the degree of risk?

Biological agents are classified into 3 categories: A, B and C, the A category includes the most dangerous biological agents.

10. Which is the latest DNA sequencing technology and what are its priorities compared to the technologies used earlier?

CRISPR-Cas9 is the latest technology that enables genome modification, through DNA sequence manipulation. This technology enables scientists to decide which genes should be expressed in plants, animals or humans; it creates the possibility of deleting unwanted features and adding desired features with a higher quality than before. This technique is revolutionary because it is cheap and simple in application.

11. Does CRISPR technology have the potential to cause damage at the gene level?

Yes. This technology has succeeded in interfering with the germ cell line and can cause inherited genetic changes across generations. Such changes can be made with negative impact. The changes can be as well controversial because they do not please everyone. Furthermore, we are not entirely sure of the effects of modified genomes on the emergence of phenotypes after 2 or 3 generations.

12. Give an example of using scientific article information for negative purposes.

In 1997, Dr. David Edwards, of Pennsylvania State University, published an article in "Science" describing a revolutionary way of delivering aerosol medication using large porous particles/molecules. The terrorists used this information to build porous particles filled with anthrax spores, allowing it to penetrate into the lungs more efficiently than ever. The same contaminated aerosols can cause transmission of pathogens that cause Q fever, flue, viral encephalitis, hemorrhagic fever, botulism and so on.

13. What are the main international regulations that control global health safety in the context of biological war?

The main regulations that control global health security in the context of biological war are: the Biological Weapon Convention (1972), the Security Council Resolution (2004) and the International Health Regulations (2005).

14. What do primary lab barriers provide?

Primary barriers keep the potentially hazardous agents inside the source (including animal cages, specialized equipment such as centrifuges or fermenters).

15. What do secondary lab barriers provide?

Secondary barriers store the agent inside the laboratory environment, in case it overcomes the primary barriers (building a specific laboratory type, directed ventilation system, waste filtration, solid and liquid waste treatment).

16. How do we divide laboratories based on the degree of the security they provide?

The laboratories are divided into: BSL-1/biosafety level one (teaching purpose), BSL-2 biosafety level 2 (primary health services, diagnostic purpose), BSL-3/biosafety level 3 (special diagnostic services and research) and BSL-4/biosafety level 4 (research using dangerous pathogenic units).

17. What types of labs are most dangerous and why?

Most dangerous to labs are the BSL-3 and the BSL-4 laboratories. The reason is that they include working with dangerous biological agents, which can cause serious human disease and can even lead to mortality. There is also the risk of transmitting disease from one individual to another, not only within the laboratory, but outdoors as well.

18. Why is it important to have safe and secure laboratories?

Safe and secure laboratories enable:

- Preservation of bio-agents that can spread infections
- Public confidence in research activities undertaken by the scientific community
- Increasing investor interest and transparency in the biomedical and biotech industry
- Preserving valuable scientific research as commercial assets
- Reducing the risk of crime and bioterrorism

19. What are the most typical routes of exposure to pathogenic agents?

- Inhalation: all laboratory processes that produce aerosols (centrifugation, mixing, fluid culture decanting etc.)

- Inoculation: through needles or sharp objects
- Ingestion: oral pipetting, food consumption in the laboratory, mouth contact with contaminated hands.

20. What are the protection means in case of a biological attack?

The protection means in case of a biological attack are: Clothes, medical protection, special masks, antibiotics and vaccines.

Activity 3. Role playing

This form of work helps the students develop strategies, skills and tools that will guide them into their professional life. Role playing also helps to identify the student's personal skills, which differentiate him from others. The students experience the effect of being in the center of attention [21].

Students play the role of bacteria and viruses. Bacteria and viruses can be used as biological agents in bioterrorism. During the role play the students on the role of bacteria and viruses meet each other for the first time. First they have to introduce themselves (to show their characteristics, the disease they cause, the organisms they affect, etc.).

Students before role play have to research and find out more about the character they will represent. To accomplish a better interpretation students and teachers may discuss about clothing or accessories that will identify the characters (T-shirts stamped with the name of the bacterium/virus or other costumes).

Teacher's role: Encourages students to be free and enjoy their role. Teacher's comments should be positive and not critical. In this way the learner will feel motivated and will adapt quickly to situations he/she has not encountered before.

Tab 1.7 Qualitative and quantitative performance evaluation criteria of role playing

Criteria	The student refuses to be part of the roleplaying, does not keep quiet and does not respect friends during the performance	The student takes on a role and tries to interact with others	The student takes on a role and engages in its interpretation accurately and seriously	The student interprets the role very good, supported by the right clothes and accessories	The student interprets very good, works to expand the dialogue enriching it with his knowledge and research on the internet, books and scientific journals
Evaluation	-	±	+	++	+++

Activity 4. Case study

Case studies are real life situations that require solutions. This method relies mainly on student participation in the discussion. Through this process they can acquire skills in critical thinking, problem solving, communication, and group interaction [22].

Scenario 1

Yes I would. Such a research is a genuine scientific work that has to be accessible from the public and appreciated by critics. Every day dozens of scientific articles are published in various journals, which increase the value of the research community and encourage the work of scientists around the world.

Scenario 2

No, I would not. Writing an article for such a research is a huge responsibility and it can lead to consequences. This information can be used by irresponsible individuals. I think a security system should be built in order to protect scientific information.

Case study 2:

At one of the depots supplying a city with water, someone is reported to have thrown a bacterial sample. Can we consider the above situation as a biological war and give the alarm for a possible bioterrorist attack?

Scenario 1

Yes, this situation can be considered as an act of biological war because someone has deliberately released a bacterial sample. The specimen may be the cause of a serious epidemic for which we may be unprepared for treatment. The consequences can be loss of human life. Giving the alarm allows all concerned structures to take appropriate action as soon as possible.

Scenario 2

It is not necessarily a bioterrorist attack. It can be fake news. The report should be first verified: is it true, what is the concerned sample, what degree of risk it represents. We cannot alert the population without understanding the situation, that's why a team of experts is created in order to analyze the situation. Even if the news is true, that bacterial sample can be negligible by mass and can be eliminated by the filtration system. It is not necessary to create panic at the population.

Case study 3:

CRISPR-Cas9 is a technology that allows scientists to modify genomes by manipulating DNA sequences. According to a report released by the director of the United States National Intelligence, it is said that a group of biologists have succeeded in interfering into the human reproductive cells. Could such activity be a concern?

Scenario 1

Not necessarily. Such advances in genetics can be used in the prevention or treatment of reproductive problems. This technology can alter the function of genes by avoiding the occurrence of a cancer as well. Genetic manipulation in the sperm or egg cell is often more effective and safer when compared to gene therapy, because it is easier to manipulate a cell than millions of cells in an organism. Parents are also given the opportunity to determine the physical characteristics of their child. The occurrence of genetic diseases can be avoided as well.

Scenario 2

Such an action is a concern because over it is difficult to control and predict the interaction of genes in the organism through different generations. If gene insertion errors occur, that error can pass to the next generation if not corrected in time or kept under control. Ethically, this technology diminishes the effect of the surprise or gift a child brings to life, because it is the parents themselves who choose the features of the child according to their desires. This changes the perception of the value of life.

Teacher's role: divides students into groups of 3-4 individuals and presents to them a problem or a situation. It listens attentively to the students reasoning, evaluates and complements students' choices. He/she guides the discussion.

Evaluation:

Tab 1.8 Qualitative and quantitative student evaluation criteria for case study performance

Criteria	The student is not included, hesitates to give his opinion on the situation that is represented to him	The student gives superficial opinions and does not defend with arguments his point of view	The student makes a judgment about the case study and defends it with arguments	The student provides a logic solution, based on what he knows and read about	The student gives a clear and logic solution, based on research; he discusses constructively with others about the solution
Evaluation	-	±	+	++	+++

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