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Regional Report

BSL-3 Laboratory User Training Program at NUITM-KEMRI

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Abstract: Pathogens handled in a Biosafety Level 3 (BSL-3) containment laboratory pose significant risks to laboratory staff and the environment. It is therefore necessary to develop competency and proficiency among laboratory workers and to promote appropriate behavior and practices that enhance safety through biosafety training. Following the installation of our BSL-3 laboratory at the Center for Microbiology Research-Kenya Medical Research Institute in 2006, a biosafety training program was developed to provide training on BSL-3 safety practices and procedures. The training program was developed based on World Health Organization specifications, with adjustments to fit our research activities and biosafety needs. The program is composed of three phases, namely initial assessment, a training phase including theory and a practicum, and a final assessment. This article reports the content of our training program.
Key words: Biosafety training program, BSL-3 laboratory, biosafety

Introduction

Emerging infectious agents, antibiotic resistant and highly infectious pathogens are important public health threats that are increasingly challenging health sectors across the world. However, manipulating such organisms presents significant risks to both laboratory workers and the environment, necessitating the application of containment strategies in order to provide a safe work environment.

Laboratory facilities are classified into four biosafety levels (BSL) based on their design features, equipment, practices and the operational procedures required when handling agents from various risk groups [1]. A BSL-1 laboratory allows for the manipulation of known microorganisms that are not likely to cause a disease and uses standard microbiological practices to provide basic containment [2]. BSL-2 laboratories are primarily diagnostic or clinical laboratories in which a broad spectrum of moderate risk microorganisms capable of causing low to severe human diseases is manipulated [1–3]. BSL-3 laboratories are medium containment laboratories that allow for work with potentially lethal microorganisms that can spread via aerosol [1]. Maximum containment is found in BSL-4 laboratories, which are designed for work with dangerous microorganisms responsible for diseases that have no known cure [1]. The general relation of risk groups to laboratory biosafety levels, practices and equipment is shown in Table 1.

The number of containment facilities, particularly level-three laboratories, has significantly increased in the recent past. This has resulted from the need to expand diagnostic and research capacity to enhance the response to the rising number of public health threats as well as a greater need to protect research personnel.

In developing countries, BSL-3 laboratories are even more necessary due to the higher disease burden characteristic of such countries. Surveillance mechanisms that manage public health may yield samples of unknown pathogenicity from time to time. BSL-3 facilities are nee-

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Table 1. Relation of risk groups to biosafety levels, practices and equipment

Risk group	Biosafety level	Laboratory type	Laboratory practices	Safety equipment
1	Basic-Biosafety Level 1	Basic teaching, research	Good microbiological techniques (GMT)	None; open bench work
2	Basic-Biosafety Level 2	Primary health services, diagnostic services, research	GMT plus protective clothing, biohazard sign	Open bench plus biological safety cabinet (BSC) for potential aerosols
3	Containment-Biosafety Level 3	Special diagnostic services, research	Level 2 plus special clothing, controlled access, directional airflow	BSC and/or other primary devices for all activities
4	Maximum containment-Biosafety Level 4	Dangerous pathogen units	Level 3 plus airlock entry, exit showers, special waste disposal	Class III BSC or positive pressure suits with class II BSCs, double-ended autoclaved (through the wall), filtered air

ded for the safe manipulation of well characterized level three agents. However, the construction of biocontainment facilities has proven to be very expensive, and the cost increases with increasing containment level [4]. This is compounded by the need for adequate maintenance budgets and qualified engineering staff. Hence, although the demand for improved diagnostic capacities and containment facilities for highly infectious agents has risen, its realization has not occurred, particularly in countries with limited resources. Such countries are also faced with numerous biosafety and biosecurity challenges, among them lack of biosafety and biosecurity awareness and poor regulation of biosafety practices [4]. Skills and knowledge necessary for safe operation in a BSL-3 laboratory are also insufficient. Despite these limitations, optimal biosafety and biosecurity practices must be adhered to while handling agents in a BSL-3 laboratory. Development of a BSL-3 laboratory user training program is therefore of utmost importance in order to increase competence and proficiency and assure safety for working in a BSL-3 laboratories.

The Nagasaki University Institute of Tropical Medicine Kenya Research Station was established in 2005 in collaboration with the Kenya Medical Research Institute (NUITM-KEMRI). Our BSL-3 laboratory was set up in 2007 at the Centre of Microbiology Research, Nairobi, Kenya [5]. Research activities involving various highly infectious agents have been carried out at the facility. These include research on highly infectious viral and bacterial agents such as arboviruses and *Mycobacterium tuberculosis*, conducted by investigators from our institution and other institutions, with some still ongoing [5]. Clinical samples from diseases outbreak in the country are also processed in the facility. In addition, the facility is used regularly to process BSL-3 agents from ongoing septicemia and virology projects.

Various strategies are applied at our laboratory to maintain biosafety and biosecurity. First, all new users have to be trained and certified before commencing any work. A mentoring and supervision program for the newly qualified personnel has also been put in place. Additionally, refresher training is conducted annually or when a new study is initiated. In-house regulations have also been developed to inform users of compulsory safety and maintenance procedures. Lastly, biosafety meetings are held at the end of every month to provide a platform for the sharing knowledge and the continuous education of users [5]. In addition, a steady supply of laboratory safety materials and Personal Protective Equipment [PPE] is maintained. Regular maintenance is also observed, alongside an annual inspection conducted by certified engineers from the NK Systems Company (Japan). These ensure the optimal functioning of the facility and hence total containment.

The Biosafety Training Program

Our biosafety training program was developed in 2007. It is implemented through an annual two-day training workshop. The program is tailor-made to suit the needs of our research activities while taking into consideration the specification recommended internationally for a biosafety training program. According to the WHO laboratory biosafety manual of 2004 [1], an effective biosafety program should incorporate the following elements;

i) Needs assessment: Determination of tasks to be carried out, and the appropriate approach.
ii) Establishing training objectives: Identification of knowledge and capabilities that a trainee is expected to demonstrate after training.
iii) Specifying training content and media: Determination of media to be used in conducting training. The media must be comprehensive and able to efficiently deliver the intended knowledge and skills.

iv) Accounting for individual learning differences: Individuals have different learning capabilities which the training program should take into account. Incorporation of different training approaches may help address this disparity.
v) Specifying learning conditions: Selection of a mode of instruction is vital in passing the required information. The adopted mode should not impede expected outcomes of the training.
vi) Training evaluation: The impact and efficiency of the training should be determined in order to establish whether the overall goal was achieved and to what extent.
vii) Training revision: Use of questionnaires and other methods to evaluate the overall training process are necessary to improve future training programs.

Our training program is composed of three phases, namely initial assessment, training phase, and final assessment. The training phase includes theory and practicum, while the assessment phases include written examinations. Training is conducted by experienced biosafety instructors from our institution and external institutions.

Initial assessment

The trainees are required to sit for a written examination before commencement of the training. The test assesses knowledge on concepts of biosafety; BSL-3 facilities; components of BSL-3 laboratories; biosafety hazards associated with the facility; measures to minimize personal harm, exposure to others and the environment; and facility maintenance. The assessment aims to determine the prior experience of the trainees, and to identify training needs.

Training Phase

The training phase is composed of theory and practicum.

a) Theory

The theoretical component of our training program is taught through lectures. The lectures are divided into three categories, namely general concepts and knowledge of biosafety and biosecurity, technical components of our BSL-3 laboratory and BSL-3 procedures. Emphasis is placed on the technical and operational aspects of our laboratory, because individual laboratories are unique and procedures applied in one may not necessarily be the same elsewhere.

Concept and knowledge

This category provides general knowledge concerning biosafety and biosecurity. Trainees are first introduced to the concept of laboratory safety and the general biosafety rules and regulations with reference to the WHO manual. Laboratory acquired infections, risk group classification of microorganisms and the principles of risk assessment are also discussed. Biosafety classification of laboratories is then described, with special emphasis on the BSL-3 laboratory and its importance in medical research. The relationship between the biosafety level of a laboratory and the risk group of biological agents handled at each level is also discussed. In addition, trainees learn how biosafety and biosecurity is achieved at each level, taking into consideration engineering controls, equipment and good laboratory practices. Lastly, trainees are introduced to KEMRI's biosafety regulations and strategies, including good laboratory practices and regulations on storage, inventory and transportation of biological agents. Lectures on topics related to mycology, virology and bacteriology are also conducted, to illustrate the application of biosafety concepts and safe handling strategies for different organisms. Such lectures are important since most of the projects implemented in our facility are centered on the above three fields.

NUITM BSL-3 laboratory: features, maintenance and management

This category provides information on the design features, maintenance, and codes of practice in our BSL-3 laboratory. Trainees are first taken through the basic design of a BSL-3 laboratory including the floor plan, physical features and operational features as well as their role in maintaining containment. Modifications and unique features of our laboratory are highlighted and discussed in terms of their functionality and how they enhance efficiency [3]. The laboratory equipment installed in the facility and the role thereof in biosafety is also discussed. The equipment includes biosafety cabinets, centrifuges and autoclaves. Secondly, trainees receive information on the facility's maintenance routine. Daily cleaning, disinfection, replacement of laboratory consumables and changing prefilters and HEPA-filters are described in detail. Thirdly, the facility's biosafety manual, which describes the biosafety rules and operations within the NUITM BSL-3 laboratory, is discussed in detail. Trainees are instructed on the do's and don'ts and codes of practice within the facility including maintenance, documentation and routine decontamination procedures. Emergency responses including spill response, facility alarms and response to accidents is also discussed. Finally, the monthly biosafety meetings are discussed. These are mandatory monthly meetings of all trained BSL-3 laboratory users currently working in the facility. The meetings offer a forum for the discussion of laboratory occurrences such as accidents, new projects, changes of procedure and any other matter of concern.

Operations inside the BSL-3 laboratory

This category covers the theoretical aspects associated with the procedure for entering, working in, and exit-

ing the BSL-3 laboratory. It was introduced to inform trainees of BSL-3 operations prior to the practical part of the training program.

The entry procedure covers instructions on the stepwise procedure of entering the BSL-3 laboratory; mainly donning PPE and passing facility check points that are useful in assessing the status of the laboratory and the anteroom. Usually our entry routine has checkpoints on the pass-box site, external door, machine room, the preparation room and the ante-room, each with a record that captures details relevant to that particular point. Laboratory procedures in the BSL-3 laboratory are then discussed, during which trainees are instructed on BSL-3 procedures, including the operation of laboratory equipment, documentation and routine maintenance procedures. Troubleshooting is also described. Finally, the procedure exiting the facility, which covers decontamination, documentation and the removal of PPE is described.

Theoretical training is conducted by biosafety trainers from our institution or professional biosafety trainers. Application lectures are conducted by experienced BLS-3 users from our institution or an external institution who have previously undertaken a research project in a BSL-3 laboratory or have an on-going project involving level-three agents. Theoretical training occupies the better part of the first day. About five lectures are presented, each with a time allocation of about thirty to forty-five minutes, with ten minutes left for discussion.

b) Practicum (demonstration and hands-on practice)

Practicum is very important in strengthening the knowledge and skills of trainees. This session is conducted through demonstration and hands-on practice, in two phases. The first phase focuses on the correct procedure of entering, working and leaving the BSL-3 laboratory. Correct donning and doffing of PPE is demonstrated first, followed by a visit to the facility check points where trainees are instructed on what to review and record. The functioning of equipment at each check point is also explained. In the BSL-3 suite, the use of laboratory equipment (biosafety cabinets, autoclave and centrifuge), decontamination, spill handling procedures and the removal of equipment or material are demonstrated. Finally, the exit procedure is discussed. The demonstration session is trainer-led. Trainees are divided into groups of four to six members. Each group spends about ten to fifteen minutes at each site.

The second phase is a trainee-led hands-on practice session covering an actual work session in the BSL-3 laboratory. In executing the work session, the trainee is expected to adhere to the recommended procedures such as donning, doffing land documentation, carrying out initial checks on the biosafety cabinet, autoclave and centrifuge to confirm availability and functioning of each piece of equipment, initial disinfection of the work surface, and arrangement of materials and equipment in the biosafety cabinet before conducting the experiment. Segregation, disinfection and removal of generated wastes should be demonstrated after completing the experiment, followed by completion of the BSL-3 laboratory leaving to-do list and exit from the laboratory.

Each work session takes about one hour and allows for both practice of the concepts learned and assessment of acquisition. The trainee executes the session based on instructions provided on a task card, with the trainer playing a supervisory role. Practical training, which takes the greater part of the second day of the training workshop, is partially integrated in lecture sessions through illustration of presentations using visual aids and demonstrations.

Final Assessment

The final assessment is accomplished through a written test similar to that used during the initial assessment. The test is based on the content covered in the entire program. Trainees who fail are reexamined after a period of two weeks but with a different set of test questions. Those who fail the second assessment are required to retake the entire training program. Completion certificates are issued upon successful completion of the training.

A discussion session is held after the evaluation to review the training program. Incorrectly answered questions in the written tests and poorly performed procedures in the practicum are discussed at length. Difficulty areas identified by the trainees are also revisited, alongside any clarifications that the trainees may need.

Discussion

At least 70 laboratory scientists have now been trained through our annual workshops. Participants mainly include new laboratory staff from our institution and laboratory scientists from other institutions. Trained BSL-3 laboratory users also attend the workshop for refresher training. Refresher training helps to maintain previously acquired skills and knowledge. It also provides an opportunity to update users on new concepts. A trainee scoring any less 70% has to undertake a supplementary examination in order to be certified. Trainees also have to demonstrate knowledge on safety practices during the hands-on session.

The structure of our program is based on the specifications provided in the WHO 2004 biosafety manual [1]. The training program is composed of an initial assessment, training phase (theory and practicum) and final assess-

ment. Tasks carried out during the training are presentations, demonstrations, discussions and trainee evaluation. These enable optimal delivery of the training content, while taking differences in learning capability into consideration. Our training objectives require a trainee to understand biosafety concepts, be able to demonstrate proficiency and correctly carry out routine BSL-3 laboratory procedures such as documentation and housekeeping by the end of the training workshop.

Our program divides theoretical knowledge into three categories to cover the major skills and knowledge necessary for working in a BSL-3 laboratory. Conceptual knowledge is a comprehensive module that covers a range of biosafety principles and practices. It recognizes the importance of theoretical knowledge in establishing consistency in safety practices. It helps the trainees to understand how biosafety and biosecurity are achieved, and how biosafety rules, regulations and practices work together to bring about personal, public and environmental safety. Given this, users are able to recognize and respond to biosafety threats and are aware of strategies to minimize biosafety risks. Such knowledge also informs their routine practices by enabling them to link an action to an outcome.

NUITM BSL-3 laboratory; features, maintenance and management category orients trainees to our laboratory. Its main focus is the physical and operational controls that inderpin biosafety. Knowledge of these aspects is useful in achieving biosafety since it enables users to distinguish normal from abnormal conditions and to apply necessary corrective measures. It also trains users on how to undertake maintenance and management of the facility, which significantly enhances containment. The third category provides instruction on how to work in the laboratory and a background for on-site demonstration and hands-on training. Prior awareness of the recommended procedures enhances active participation in practical training sessions, which in turm improves understanding and the acquisition of practical concepts.

Practicum provides instruction on correct procedures for each activity and procedure. Demonstration is conducted by experienced biosafety trainers. The trainees thereafter conduct a hands-on practice session under the supervision of a trainer. Practical sessions not only enhance skill and competence but also provide an opportunity for clarification of difficult areas and assessment of safety habits.

Training evaluation is by written test and partially via on-site evaluation during the hands-on session. Training revision is achieved using a self-administered questionnaire by which the suitability of the training program is gauged. The opinions of trainees on the entire training workshop are also collected for purposes of continuously improvement of the program. Mentorship follows completion of the training program. A newly trained BSL-3 laboratory user is assigned a supervisor whose role is to oversee the trainee's BSL-3 sessions until he or she has demonstrated sufficient proficiency in BSL-3 procedures and practices. During this period, the trainee is introduced to work with live organisms as well as carrying out routine BSL-3 laboratory maintenance procedures. An ideal mentor is usually a trained and experienced laboratory staff member or biosafety officer, and the duration of mentorship depends on the new user's speed in mastering recommended practices and procedures.

During the first three years of administering our training program, focus was placed mainly on the theoretical aspects of biosafety and biosecurity, with a limited practical training component. The hands-on training component was later included and the program schedule reorganized to increase the time allocated for practicum to two, two-hour long sessions. The lecture content was also revised to increase coverage of practical concepts that enhance safety in our facility.

Our training program integrates theoretical training with practical training to provide the knowledge and skills necessary for ensuring safe operations within a BSL-3 laboratory. Our approach builds on the theoretical knowledge by enhancing practical skills and uses both aspects to promote adherence to biosafety guidelines. Collectively, these enhance competency and proficiency among BSL-3 laboratory users and ensure adherence to biosafety and biosecurity regulations.

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