

Collection Activities in the Field of Use of Pathogenic Microorganisms in Ensuring Biological Safety of the Russian Federation

*Yakhita Soltanovna**

Kadyrov Chechen State University, Grozny, Russia

Abstract. This review reflects the current state of collection activities related to the use of pathogenic microorganisms. A modern regulatory framework regulating key aspects and regulating collection activities, which is a critical step in microbial collection development, is presented. A number of key tasks facing state collections and solutions to ensure biosafety and sanitary-epidemiological well-being of the population of the Russian Federation have been defined. Directions for improving the state collection of pathogenic microorganisms are formulated.

1 Introduction

Pathogenic microbial systems play an important role in a number of measures to ensure the sanitary and epidemiological well-being of the population. In the case of natural local infection, research is needed on the influx and spread of pathogens in atypical areas and their characteristics when monitoring the area. The most important direction is also the timely development of diagnostic and preventive drugs against pathogens of dangerous infectious diseases. The development of such drugs is preceded by a detailed study of the properties of pathogens circulating in the territory of interest to the researcher in a certain period of time, which is necessary for understanding the directions for designing test systems, vaccines, various diagnostic tools, etc. In addition, the availability of viable samples of pathogenic strains in most cases depends on the stages of creating such drugs, which can be based on various components obtained from microbial cells. Collections of pathogenic strains are necessary for testing designed test systems both as a control, to assess the specificity and sensitivity of the drug, and in the form of heterologous analogues. The preservation of pathogenic microorganisms is carried out by a set of measures to ensure collection activities [1].

In the Russian Federation, this function is assigned to State collections of pathogenic microorganisms. This review reflects the stages of creating collections of pathogenic microorganisms, the regulatory framework governing their activities at the domestic and international levels, as well as the main tasks facing collections in the modern period, approaches to their solution and development prospects. The formation of microbial collections is an important step in the development of microbiology. The first collection of

* Corresponding author: xabieva.zaira@bk.ru

“living” culture was created in Prague in 1890 by Frantisek Kralj at the Medical Faculty of the Institute of Hygiene. It contained hundreds of cultures and arrangements fixed to glass slides, most of which were later exported to the United States and included in the American Type Culture Collection [2]. The oldest existing collections of microorganisms are the two mycological collections. One of them was founded in Belgium in 1894, the Mushroom Collection at the Catholic University of Leuven. A few years later, in the Netherlands in 1906, the Central Bureau of Mycological Collections [Collection of the Central Bureau of Mycological Cultures (CBS), Utrecht, The Netherlands] was founded. In the early 20th century, numerous collections of microorganisms were created by individual European researchers and scientific organizations, but all of them, as well as those created in Japan in 1884, are now lost [3].

In 1920, the National Type Culture Collection (NCTC), containing more than 5,000 bacterial strains, was created in the UK. Founded five years later in the United States, the American Type Culture Collection (ATCC) is now considered the world's largest biological resource center for the storage and study of microorganisms. The collection contains 18,000 bacterial strains belonging to 750 genera. ATCC has more than 3,600 type strains that form the basis of bacterial taxonomic research. The formation of microbial collections in Russia began in the 1930s and their organization reached its peak in the 1950s and 1960s, when a total of 36 collections were formed [4]. One of the first collections of bacterial pathogenic microorganisms with officially fixed status (Order No. 205/4/30, 1955 of the Ministry of Health of the USSR) is the collection of pathogenic microorganisms of the I-II pathogenic group. Russian Research Anti-Plague Institute “Microbe” and III-IV Pathogenic Group - State Institute of Standardization and Control. L. A. Tarasevich. Under this order, the official status of the collection was assigned to the Museum of Life and Culture of the Institute of Epidemiology and Microbiology. Virus Research Institute Gamaleya. Ivanovsky, Irkutsk Anti-Plague Institute and many other institutions. Currently, the most important collection of microorganisms of pathogenic Groups I-II in the Russian Federation is located in the Russian Academy of Sciences, an agency under the Federal Service for the protection of consumer rights and supervision of human welfare, Ministry of Defense. Ministry of Agriculture of the Russian Federation and the Russian Federation.

2 Research Methodology

Depending on the structure, function and scope of the work performed, all collections of pathogenic microorganisms operating on the territory of the Russian Federation can be divided into three types: state, institutional and work. The most important is the modern center, which provides storage from simple storage of biomaterials, and the state collection, which extends its functionality using a comprehensive research and collection system using modern methods. The creation of such collections is regulated by the orders of ministries and departments.

In addition, the collection of the condition of pathogenic microorganisms solves the fundamental problem that aims to:

- Perform taxonomic studies to identify and comprehensively analyze new species and new pathogenic variants formed within the species.
- Clarify the taxonomic position of cultures with atypical phenotypes and genotypes.
- Certification of microbiology, molecular genetics and epidemiology of natural and genetically modified microbial systems.

As part of the activities of research organizations engaged in the study of pathogenic microorganisms, institutional collections are often created. They are separate divisions of institutions licensed to work with pathogenic microorganisms. Creation of institutional collections is regulated by orders of the head of the organization. The main functions of

institutional collections are: vformation, replenishment, maintenance and accounting of the collection fund of pathogenic microorganisms of those nosology's that are handled in this organization; vstorage of the collection fund in a freeze-dried state, in conditions of low-temperature freezing and cryopreservation; vimprovement of storage methods for pathogenic microorganisms; vproviding departments of the institution with high-quality samples of microbial strains; v maintaining databases and catalogues of collection strains. If a limited number of pathogenic microorganisms are used in the scientific and practical activities of the organization, their working collections are created. Working collections should be understood as sets of strains that are in safe custody of employees working with them in departments that have a sanitary and epidemiological conclusion on the right to carry out activities with pathogenic microorganisms. The creation of working collections is carried out with the permission of the heads of the institution and division. The main functions of working collections are: vcarrying out research, diagnostic, production and other activities related to the use of pathogenic microorganisms; vmaintaining the working collection strains in a viable state.

3 Results and discussions

The basis of the collection fund when it is formed is usually a set of typical, neotypic and reference strains of microorganisms of that specific affiliation in the field of activity with which the collection is allowed. The Bacteriological Code refers to the type strain on which the author, who first described the organism he named, based his description of the latter, which he or a subsequent author definitely designated as a type. At the same time, a viable culture of a typical strain should be placed in at least two specialized collections located in different countries, with the possibility of freely obtaining subculture. The neotype is considered a strain that is accepted instead of the standard one, in case of loss of the latter. Authors proposing a strain as an aneotype should provide detailed information about its properties, the availability of viable samples in authorized collections and evidence of loss of the type strain, as well as that the proposed neotype agrees well with the description of the original one. voltage. Reference cultures usually include strains that are neither typical nor non-typical, but are used as reference strains for taxonomic, serological, chemical or other types of analysis. Such a microorganism should be identified, at least up to the genus and species, catalogued and described by its characteristics.

In addition, based on the tasks performed by each individual collection center, its fund may include strains differentiated by purpose and origin:

- natural strains of pathogens of particularly dangerous infections isolated from sick people, carriers, animals and from environmental objects characterizing epidemic and epizootic manifestations, interepidemic periods, allowing to obtain the idea of the circulation of pathogens in certain areas for a certain period of time;
- natural and genetically modified strains of microorganisms of pathogenicity groups I–IV, deposited in the order of copyright and patent;
- microbial production strains of pathogenic groups I-II, used in the production of diagnostic and preventive drugs;
- reference test strains of microorganisms of pathogenicity groups I-IV, used in quality control of nutrient media in medical practice;
- vaccine strains of pathogens of plague, anthrax, tularemia and brucellosis;
- training strains – strains used in the educational process that mimic strains.

In addition, canning by this method provides greater stability for many microorganisms, especially in comparison with other methods of drying or periodic transplantation. In addition, due to the characteristics of the freeze-drying procedure, unlike many other

canning methods, a large number of containers can be obtained and stored for each culture, which improves the safety of collected funds and allows transportation of microbial strains over a significant distance at a minimum cost. [5].

Another option for long-term storage of microbial cells in a viable state is their maintenance at low temperatures in the presence of cry protective media in refrigeration units. Most bacteria are able to survive at temperatures below -60 °C, while maintaining a high concentration of living cells for a long period of time. This approach is easiest to implement compared to other methods of long-term preservation of microorganisms. The implementation of the low-temperature freezing process is characterized by minimal preparatory work, preservation of stored materials and rapid implementation of the extraction stage. Frozen samples do not undergo any genetic changes and always retain their original characteristics [14]. In addition, some cultures are characterized by higher cell viability by this canning method than by drying or freeze-drying. The biggest vulnerabilities of low-temperature canning methods are the need to ensure uninterrupted operation of refrigeration units and the complexity of transporting frozen materials.

One of the variants of cryopreservation is cryopreservation, a method characterized by storing substances in liquid nitrogen. This approach is designed for long-term preservation of biological materials. Depending on the temperature conditions that do not allow thawing of objects, the titers of living cells practically do not change, so microbial cultures can be stored for almost unlimited time [15]. The most reliable way to keep the temperature of the stored sample constant is to dip the sample directly into liquid nitrogen, which ensures a storage mode at -196 °C. However, this procedure risks contaminating containers and storage rooms with pathogenic biological agents due to the possibility of liquid nitrogen entering the cry probe [6].

As a result, it is recommended that toxic microorganisms be stored in liquid nitrogen vapor and that the container does not come into contact with liquid gases. It should be noted that there is no universal approach to cryopreservation of various types of microorganisms that require the use of various cryoprotectors and the freezing rate of cell suspensions. Nevertheless, cryopreservation is convenient for maintaining microorganisms that do not withstand the stress conditions of other methods, and a variety of cryopreservation equipment is currently available [8]. The most important task of the collection is to determine the definition of the taxonomic series and compliance with the reported passport data. Initially, the study of this direction was based on cultural and morphological, phenotypic and serological methods. Further steps in this direction have been taken with the development of a methodology aimed at the study of the genome of collection strains [13].

Polymerase chains are widely used reactions (PCR), which are used to identify the main genetic determinants of toxicity, pathogenicity and various species-specific markers. To date, numerous molecular genetic methods have been developed for classifying microbial lineages and determining their taxonomic affiliation. Since a universal approach to accurately determine the systematic location of the studied culture has not yet been established, it is necessary to use identification algorithms based on two or three approaches that complement each other. Therefore, the general approach to the designation of pathogen types for infectious diseases, cholera, yatosis, anthrax, brucellosis, Glander and melioidosis is multi - focal sequencing (MLST-from English. Multilocus sequence typing), ribotyping, and multi-position VNTR analysis [7].

The most important task of the collection activity is to determine the taxonomic affiliation of the strain and ensure that it complies with the reported passport data. Initially, the study of this direction was based on cultural-morphological, phenotypic and serological methods. Further steps in this direction have been taken with the development of technologies aimed at the study of the genome of collected strains. Polymerase chain

reaction (PCR) is a widely used reaction (PCR) to identify major genetic determinants such as toxicity and pathogenicity and various species-specific markers.

To date, numerous molecular genetic methods have been developed for classifying microbial lineages and determining their taxonomic affiliation. Since a universal approach to accurately determine the systematic location of the studied culture has not yet been established, it is necessary to use algorithms for identification based on two or three approaches that complement each other [12]. Therefore, common approaches to classify pathogens of plague, cholera, yatosis, anthrax, brucellosis, Glander and melioidosis are multifocal sequencing, ribotyping, and multi-position VNTR analysis.

This method is based on limited intercept length polymorphism, is reproducible and has been proven to be a way to distinguish cholera strains based on their origin in various cholera endemic areas. A practical example of the molecular type is the investigation of the source of the mail terrorism incident in the United States in 2001. Collection variant set of pathogen anthrax, where you can set the variant, source, and author names of anthrax spore distribution [9]. To establish the authenticity and systematic position of collection variants, we have firmly entered the functional system of foreign collections by using various molecular type systems and creating algorithms based on them. This package provides the ability to connect abstract callbacks to class methods, functions, or function objects, and includes an adapter class for connecting other callbacks.

In Germany, microbial and cell culture collection, riboprinting and gene-gene hybridization are used to establish the authenticity of bacterial strains. Russia's modern approach to establishing the systematic location and authenticity of strains has been implemented in many non-medical collections. Recently, this approach has begun to be used in collections supporting pathogenic microbial strains [10].

At the same time, the development of integrated algorithms to determine taxonomic location and establish the reliability of collected strains remains one of the priorities of current national pathogenic microbial collection. In this regard, research and development activities to solve these problems are planned and implemented within the framework of the Federal Goal program "State System of Chemical and Biological Safety (2015-2017)".

4 Conclusions

Therefore, collection activities related to the use of pathogenic microorganisms are the direction of scientific activity to ensure the biological safety, hygiene and epidemiological well-being of the population of the Russian Federation, successfully implemented and harmoniously developed. However, some aspects of this important feature require further improvement.

First of all, this is due to the development and implementation of an integrated approach to the functioning and implementation principles of the main mission of the national collection of pathogenic microorganisms, provided by the integrated regulations on collection activities in the field of I pathogens. It is also necessary to develop a single document for all collections to update the regulatory and methodological framework governing the rules for depositing pathogenic microbial strains. An important task facing state collections is the creation of a unified panel of reference strains of pathogenic microorganisms, which is necessary to ensure research, diagnostic, production and educational activities. In addition, it is necessary to constantly coordinate the processes of improving the methods of molecular typing of pathogens of especially dangerous infections, their conservation, storage and information support, taking into account the emergence of new approaches, methods and technologies, maintaining them at the modern world level.

References

1. J. R. Porter, The role of culture collections in the era molecular biology. ATCC 50th Anniversary Symposium, American Society for Microbiology, 62-72. (Washington, DC, 2020).
2. Y. Tsunematsu, Japanese Federation of Culture Collections. Proc. Int. Conference on Culture Collections, 79-83 (2020).
3. D. T. Dzhatdoeva, Distribution of species of medicinal plants of the flora of Karachay-Cherkessia by floristic regions and high-altitude zones, **2**, 68-78 (2012).
4. D. T. Dzhatdoeva, Geographical analysis of the flora of medicinal plants of Karachay-Cherkessia, **1**, 33-36 (2012).
5. C. Cagnin, E. Amanatidou, M. Keenan, Orienting European Innovation Systems towards Grand Challenges and the Roles that FTA Can Play, **39(2)**, 140-152 (2020).
6. H. P. Seeliger, The Medical Culture Collection, Impact of Science on Society, **158**, 167-174 (2020).
7. M. J. Ashwood-Smith, *Low Temperature Preservation in Medicine and Biology*, 323-327 (Tunbridge, Wells: Pitman Medical, 2020).
8. L. F. Gibson, J. T. Khouri, Storage and survival of bacteria by ultrafreeze, Lett. Appl. Microbiol., **3**, 127-129 (2020).
9. D. Fountain, M. Ralston, Net al. Liquid nitrogen freezers: a potential source of microbial contamination of hematopoietic stem cell components, Transfusion, **37**, 585-591 (2018).
10. B. W. W. Grout, G. J. Morris, Contaminated liquid nitrogen vapour as a risk factor in pathogen transfer, Theriogenology, **71**, 1079-1082 (2019).
11. A. E. Hawkins, M. A. Zuckerman, R. J. Gilson et al., Hepatitis B nucleotide sequence analysis: Linking an outbreak of acute Hepatitis B to contamination of a cryopreservation tank, J. Virol. Methods, **60**, 81-88 (2020).
12. R. Kh. Ilyasov, Spline modeling and analysis of relationships in the economy with the possible presence of regression switching points, **11(4)**, 165-175 (2018).
13. K. M.-S. Murtazova Ecological and economic assessment of sectoral agricultural technologies, **3(15)**, 68-71 (2021).
14. I. U. Shakhgiraev, Green structures in the concept of sustainable development of modern cities. Regional building complex: investment practice and implementation of PPP: materials of the All-Russian scientific and practical conference, 376-383 (Moscow, 2019).
15. I. U. Shakhgiraev, Management of the financial security of an enterprise, IV All-Russian Scientific and Practical Conference “The Impact of the New Geopolitical Reality on Public Administration” and development of the Russian Federation”, 236-242 (Grozny, 2021).