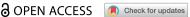




## REVIEW ARTICLE



# Strengthening the Korean Network of Microbial Culture Collections in the Microbiome Era

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#### **ABSTRACT**

Microbes are critical contributors in main areas of biotechnology, including green, red, and white biotechnology. This is why the importance of the preservation of microbial resources cannot be emphasized enough. Culture collections are repositories not only for the preservation and maintenance of a large variety of microbial resources and the associated data but also for their distribution in a quality-controlled manner. The mission of culture collections facilitates and supports utilization of microbial resources for research, education, and industrial purposes. Led by the World Federation for Culture Collections, an international organization committed to fostering the activities of culture collections of microorganisms and cell lines, more than 850 culture collections from 80 countries and regions work together to ensure the perpetuation of microbial resources. In addition, domestic networks, such as Japan Society for Culture Collections and United States Culture Collection Network along with regional networks for Europe, Latin America, and Asia thrive to ensure the long-term viability of microbial resources. The Republic of Korea recently took the first step in networking through the coordination of six ministries which house nine national microbial culture collections. With an explosion in microbiome research and a dramatic increase in the number of microbiome samples, the considerable challenge of culture collections will therefore be implementing the biobanking infrastructure of microbiome samples. Creating a domestic network of national culture collections is a key factor in efficiently and comprehensively managing nation-led microbiome research projects, particularly resulting microbiome samples. In this context, this review aims to provide an overview of microbial culture collection network and their future role to address the challenge in the microbiome era.

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## 1. Introduction

Exploitation of natural resources and environmental disturbances cause severe problems in promoting sustainable development goals of the United Nations. Microbes are recognized as the indispensable resources to solve these challenges. Indeed, they have been extensively utilized in various fields, including agriculture, food, health, and waste management over the past five decades [1-4]. Microbial-derived products are integral to our daily lives for both food and non-food domains, such as bread, cheese, wine, probiotics, and so forth. The constant demand for microbes in a wide range of applications stresses the necessity for long-term preservation of these valuable resources. Therefore, culture collections play a vital role as biological banks, as they preserve different types of microbial resources and provide high quality resources to the stakeholders for research, education, and industrial purposes [5-10].

The wide application of microbes has been accelerated due to technological advances and a better understanding of microbial diversity and functional potential. In particular, the technological advances mainly because of the rapid development of sequencing technologies and computational tools have allowed an exponential rise in microbiome studies, producing an increasing number of microbiome samples. These include intact samples, pure cultures, and mixed microbiomes. There are quite a few reasons why preservation of microbiome samples is imperative, but the most important is to prepare for shrinking of microbial diversity of clinical, environmental, and industrial importance. Hence, one of the challenges culture collections faces is the development of optimized methodologies for the preservation of microbiome samples and the assessment of preservation's success in terms of maintaining microbial composition and functionality [11].

With the common aim of promoting extensive utilization of microbial resources by their maintenance and distribution, culture collections around the world collaborate through an international body, namely World Federation for Culture Collections (WFCC). At the 15th International Conference on Culture Collections (ICCC15), the most recent WFCC meeting held on June 2023 in Portugal, the need for the biobanking infrastructure of microbiome samples was discussed. Members of WFCC acknowledged during the discussion that culture collections should take the lead on this task and collaboration is inevitable to move forward. Aside from WFCC, transnational networks developed at regional/ continental level or theme-centered networks, such as the Microbiota Vault (https://www.microbiotavault. org), also play a significant role for the sustainable use of microbial diversity and its conservation. Importantly, maintaining and improving a close liaison between culture collections at the domestic level is an essential prerequisite for creating a cooperative network more widely. In this context, this review highlights (i) the current status and importance of culture collection network in the world, (ii) efforts for developing a domestic network of national culture collections in Korea, and (iii) role of the Korean network of national culture collections in the microbiome era.

# 2. Networks of microbial culture collections in the world

Since the first culture collection was established by Professor Frantisek Král in Prague in about 1890, other culture collections were created such as the Westerdijk Fungal Biodiversity Institute (KNAWWI, formerly known as Centraalbureau voor Schimmelcultures, KNAW-CBS) in 1906 which is specialized for fungi and the American Type Culture Collection (ATCC) in 1925 for different types of microorganisms [10]. Over a more than a century

old history, the mission of culture collections is the collection, preservation, and distribution of microbial resources together with the associated data [12]. This mission can be achieved by services and expertise related to conservation/quality assurance, taxonomic classification/identification, and management of microbial resources. Investment into valuable microbial resources through establishment of culture collections and related services enables the conservation and sustainable use of microbial diversity, which further facilitates microbial research and industrial development. To achieve a synergistic effect, many countries, mostly highly developed countries, put great emphasis on improving interactions between culture collections and their users. Some examples include UK National Culture Collection (UKNCC), Japan Federation of Culture Collections (JFCC), and United States Culture Collection Network (USCCN).

The UK's network of national culture collections was established in 1947 (http://www.ukncc.co.uk). UKNCC coordinates activities of the member collections among scientists and sharing of technical knowledge and best practices. While maintaining close relationships among nine national collections, UKNCC offers deposit, preservation, identification, and characterization services via its online database. JFCC was initiated by the Japanese culture collections in 1951 and is presently known as Japan Society for Culture Collections (JSCC) [13]. JSCC is one of the well-known and the most successful examples of domestic collaboration networks, as it was later launched into Japan Society for Microbial Resources and Systematics (JSMRS) in collaboration with Japan Society for Microbial Systematics (JSMS) (https://www.jsmrs.jp). The new society contributes not only to the conservation and sustainable use of microbial resources but also to promotion of scientific research and technologies across different areas of microbiology. Formed in 2012, USCCN brings together culture collection community in the United States to support the safe and responsible utilization of microbial resources [14]. While USCCN sought to develop collaborations and promote interactions among culture collection curators through meetings, site visits, and joint publications during the first phase (2012-2020), in the current second phase the network aims to expand the scope by engaging scientists across multiple disciplines to address technical challenges common to all culture collections (https://usccn.org).

Starting from the domestic level, countries have expanded cooperative relations with other countries at the regional/continental level, some of which include European Culture Collections' Organization (ECCO, 1981), Latin American Federation for Culture Collections (FELACC, 2004), and Asian Consortium for the Conservation and Sustainable Use of Microbial Resources (ACM, 2004). Toward the global cooperation on microbial resources, WFCC plays a major role, bringing together more than 850 culture collections from 80 countries and regions. As the data center of WFCC, World Data Center of Microorganisms (WDCM, https://www. wdcm.org) was established almost 60 years ago and offers culture collection catalogues, services, and molecular data concerned with culture collections [12].

Despite a century-long history and the significant contribution of culture collections to conserve microbial diversity, they have not been given sufficient attention until fairly recently. In accordance with the enactment of the Nagoya Protocol in 2014 and the enormous growth of bioindustry, maintenance and utilization of microbial resources began to gain more attention. The Protocol prompted scientists and decision-makers to improve the quality of microbial resources and reduce their dependence on overseas resources. This has brought the practical need to establish and operate microbial culture collections around the world for the preservation and distribution of promising microbial resources. At the time of writing (June 2024), there are 857 culture collections in 80 countries and regions registered with WDCM database, of which 315 are in Asia, 271 in Europe, 213 in America, 42 in Oceania, and 27 in Africa (https://ccinfo.wdcm.org). When looking at a total of 4,012,947 strains by types of microorganisms, bacteria are 1,613,325, fungi are 861,992, 39,858 are viruses, and 32,629 are cell lines (https://ccinfo. wdcm.org on June 2024). Although Asia has the biggest number of culture collections, most fungal species are represented in European culture collections due to the capability of KNAW-WI, as one of the World's leading authorities in mycology, containing over 100,000 living strains of fungi (https://wi.knaw. nl; 15]. Referring to the data presented by Sharma and Shouche in 2014 (647 culture collections in 70 countries), the number of culture collections has indeed increased significantly in the last 10 years. Some important culture collections are listed in Table 1. Data derived from WDCM revealed that Asian countries particularly three countries of East Asia, including China, Japan, and Republic of Korea, have occupied higher ranks in the number of microbial strains, accounting for 26.3% and 34.9% in 2014 and 2024, respectively. It is interesting to note that Brazil has the highest number of microbial culture collections registered with WDCM in both 2014 and 2024. The Brazilian government gives further impetus to culture collection management to strengthen the preservation of Brazilian biodiversity in different biomes and support the bio-economy [17].

# 3. Efforts for developing a domestic network of national culture collections in Korea

Currently, a total of 27 microbial culture collections, including Korean Agricultural Culture Collection (KACC), Korean Collection for Type Cultures (KCTC), Korean Culture Collection of Aquatic Microorganisms (KoCAM), and Korean Veterinary Culture Collection (KVCC) are registered with

Table 1. Microbial culture collections and their holdings of top 20 countries. Different colors represent different continents; blue for Asia, green for Oceania, yellow for America, and white for Europe.

	2014*			2024**			
		No. of			No. of		
Rank	Country	collections	No. of holdings	Country	collections	No. of holdings	
1	Japan	25	247,037	China	52	920,101	
2	UŠA	24	242,436	USA	37	343,835	
3	Brazil	65	176,902	Belgium	7	296,555	
4	India	27	152,849	Japan	26	265,863	
5	China	25	146,162	India	34	225,072	
5	Korea	21	145,009	Korea	27	213,561	
7	Germany	13	93,368	Chinese Taipei	3	158,143	
3	Netherlands	6	90,775	Brazil	91	137,675	
9	Denmark	3	88,566	Iran	24	116,754	
10	France	38	86,350	Denmark	3	112,066	
11	U.K.	19	84,210	Netherlands	6	110,775	
12	Astralia	34	82,946	Germany	15	107,443	
13	Canada	18	82,315	France	38	103,095	
14	Chinese Taipei	2	67,227	Australia	34	97,207	
15	Russia	22	60,168	Canada	20	88,741	
16	Belgium	7	56,128	U.K.	19	87,669	
17	Sweden	3	52,700	Russia	30	74,104	
18	Thailand	60	43,251	Italy	25	62,195	
19	New Zealand	6	25,045	Sweden	4	53,020	
20	Italy	10	23,879	Thailand	65	43,687	

<sup>\*</sup>Data from [16].

<sup>\*\*</sup>Data from https://www.wfcc.info (April 2024).

WDCM database, making the Republic of Korea top 6 in the world in the number of microbial strains (213,561 strains, 5.3% of total microbial strains). These culture collections are supported by different bodies, of which 17 collections are supported by universities while eight collections are by governmental or semi-governmental organizations and the remaining collections are by private or industries. This is consistent with the global pattern across countries and regions where a large percentage of culture collections are supported by universities (43.8%) followed by governmental organizations (37.9%) (https://www.wfcc.info). Despite the considerable number of Korean culture collections and microbial strains, there has not been an official or unofficial venue bringing together all culture collections. In 2021, the Korean government initiated a multi-ministry project to efficiently manage biological resources and the associated data with an aim of accelerating the growth of bioresearch. Specifically, a total of 274 scattered biological resource banks have been reorganized into 14 themed clusters through this project. Among the 14 clusters, the microbial cluster is led by two designated hub banks, KACC of the Rural Development Administration and KCTC of the Ministry of Science and ICT (MSIT), specializing their roles in the respective fields, green and red biotechnology, respectively. However, due to the wide applicability of microbes, not all microbial culture collections are organized into the same microbial cluster, but instead spread into other four clusters, including aquatic life, marine life, pathogen, and wildlife clusters. This is contradictory to the ultimate objective of the Korean government's multi-ministry project, which strives to put together scattered biological resource banks into the same cluster.

To develop a domestic collaboration network in the Republic of Korea for the first time, representatives of the nine national culture collections belonging to six ministries came together in July 2023 to discuss how culture collections can better serve users and stakeholders. They showcased the history and existing resources available in the culture collections (Table 2). Briefly, the culture collections preserve various types of microorganisms, including bacteria, fungi, viruses, algae, and cell lines, with bacterial strains accounting for the highest proportion compared to other types. In addition, some culture collections, such as KACC, KCTC, and the Wildlife Biological Resources Bank preserve a significant proportion of fungal strains. The meeting participants shared ideas specifically on ways to improve interactions between culture collections to advance the common goal of furthering education, research, and technological development. Some of the main and common challenges in most of the culture collections are lack of proper infrastructures and funds as well as trained personnel. Recurring themes during the meeting include how to build sustainable

**Table 2.** Korean national microbial culture collections and existing resources as of December 2023. Culture collections are listed in alphabetical order.

Culture collection	Acronym	Governmental body	Years established	Holdings
Bank of Bioresources from Island and Coast	BOBIC	Ministry of Environment	2021	Total 4,333 (Bacteria/Archaea 4,060; Algae 124; Fungi 149)
Freshwater Bioresources Culture Collection	FBCC	Ministry of Environment	2016	Total 21,124 (Bacteria/ Archaea 15,849; Fungi 3,725; Algae 1,550)
Korean Agricultural Culture Collection	KACC	Rural Development Administration	1995	Total 26,562 (Bacteria 13,104; Molds 9,002 ; Mushroom 4,867; Yeasts 611)
Korean Culture collection of Aquatic Microorganisms	KoCAM	Ministry of Oceans and Fisheries	2002	Total 6,843 (Bacteria 6,843)
Korean Collection for Type Culture	КСТС	Minstry of Science and ICT	1985	Total 35,524 (Bacteria/ Archaea 23,838 ; Molds 5,541; Yeasts 3,073; Microalgae 1,887 ; Plant cell lines 962 ; Animal cell lines 223)
Korean Veterinary Culture Collection	KVCC	Minsitry of Agricultrue, Food and Rural Affairs	2008	Total 10,591 (Bacteria 8,347; Antiserum 322; Virus 987; Plasmid 141: etc)
Marine BioBank (National Marine Biodiversity Institute of Korea)	MABIK	Ministry of Oceans and Fisheries	2019	Total 9,192 (Bacteria 8,037; Fungi 936; Microalgae 219)
National Culture Collection for Pathogen	NCCP	Korea Disease Control and Prevention Agency	2004	Total 7,944 (Bacteria 6,395; Fungi 716; Virus 519; Derivatives 314)
Wildlife Biological Resources Bank	NIBR Biobank	Ministry of Environment	2010	Total 24,802 (Bacteria 12,347; Fungi 11,535; Algae 920)

network for preservation and quality control of microbial resources, some of which are summarized below.

- I. The first step toward better dealing with the Convention on Biological Diversity and the Nagoya Protocol is to enhance a network among the domestic culture collections across organizational and functional boundaries.
- II. Preservation facilities and methods are very important to secure microbial resources and ensure their quality, and cooperation can promote best practice sharing and accelerate technological development.
- III. With the increasing demand of the users for microbial resources in the bio-economy era, it is necessary to establish a joint strategic plan to secure manpower and long-term core funding.
- IV. Korean microbial culture collections need to shoulder more responsibilities in the internacommunities, tional including **ACM** and WFCC.
- V. The participating culture collections communicate for the regular sharing of information and ideas to have a common voice in discussions of curation and management of culture collections via at least an annual meeting.

# 4. Future role of the Korean network of national culture collections in the microbiome era

Since the publication of a study that provided evidence that human obese phenotype could be transferred to germ-free mice via fecal microbiome transplantation [18], microbiome research has evolved rapidly and become a popular topic in both the scientific community and the public [19,20]. This has resulted in thousands of publications and remarkable research investment across numerous fields of science, including agriculture and food domain. Microbiome-related studies are gaining more and more attention, as they provide a comprehensive view of microbial structure and function of any environmental or clinical samples at a given point in time. The human gut microbiome and its role in health and disease are one of the best examples of how microbiome-related studies help us understand the structure and function of microbial communities in a better way. Another good example is the microbiome inhabiting the soil and plant and its role in plant growth and health enhancement toward sustainable agriculture. Furthermore, increasing evidence shows that marine microbiome is important for biotechnological applications as well as biogeochemical function of the ocean [21,22].

In response to the increasingly recognized potential of microbiome, the Korean government has been keen to encourage microbiome research to enhance national industries. In September 2020, a coordinated effort was made across ten government ministries to finalize a "Plan to Promote Green-Bio Convergence Emerging Industry". The plan was developed to double the size of Korea's green biotechnology industry by 2030 with the focus on five promising areas including microbiome. In early 2023, the Ministry of Agriculture, Food and Rural Affairs (MAFRA) announced "Green Bio Industry Promotion Strategy" to foster Korea's agricultural competitiveness in the world market with the R&D support of core technologies again including microbiome. As a follow-up on this strategy, MAFRA announced an upcoming call for proposals for research projects over the five-year period (2024-2028). Consisting of a total of seven projects, the goal of the proposals is to preoccupy core technologies for the microbiome as the key to next generation bioindustry. Transiting from an example of agriculture to human health sector, the Ministry of Health and Welfare together with the Korea Disease Control and Prevention Agency support a five-year project (2023-2027) to establish a national-level platform for human microbiome research specialized in incurable diseases and develop microbiome-targeted therapy. Although focuses limited to human microbiome, this project plans to create standardized guidelines and a framework necessary for conducting microbiome research from sample collection to data analysis. Another five-year project, which was initiated by MSIT in 2023, is promoting the development of source technologies for microbiome-based treatment of incurable diseases and anti-cancer. Apart from agriculture and human health sectors, the Ministry of Oceans and Fisheries supports marine microbiome research (2021-2026) for improvement of control technologies of infectious viruses in the marine environment, development of marine probiotics, and support for commercialization of functional materials derived from seaweed.

Despite the significance of microbiomes in human and ecosystem health, unfortunately, the microbial niches and diversity continue to be threatened by urbanization, industrialization, and climate change [23]. Failure to preserve and cultivate these valuable microbial resources may result in shrinkage of microbial diversity of clinical, environmental, and industrial importance. Taking into consideration the development of novel cultivation techniques for uncultured microbes, preservation of intact samples

and mixed microbiomes is imperative for future cultivation. Except for some foodborne pathogens, such as Campylobacter species whose lyophilization procedure has been recently developed for long-term preservation [24], it is considered that preservation methods for purified microbial isolates are almost optimized. On the other hand, there has been relatively little work focused on preservation of co-cultures or whole microbiomes [25]. Therefore, the key question that needs to be answered is how to assess proper sample preservation methods for microbiome studies and their functional potential beyond individual microbial isolates, as different microbes need different preservation methods. Attempts have been made toward the preservation of microbiomes, most of which targeted fecal or soil samples. For fecal samples, the protocols for processing and preservation are recognized to be successestablished, and the fecal microbiome transplantation has already been under practice for improved human health [26].

Even with the progress in developing methodological details, there is currently no standardized protocol for preservation of soil microbiomes. Although further considerations specific to research objectives should be given to sample storage methods, freezing at -20°C or -80°C is generally considered the gold standard for long-term preservation of soil microbiomes to limit microbial activity or proliferation [27]. One of the most recent studies found negligible effects on soil microbiome composition between samples preserved at various conditions in the short-term experiment with changes observed only in beta diversity of microbiome in the long-term experiment [28]. This finding is consistent with other previous studies where soil microbial communities are relatively robust to different preservation methods [29,30] except for rare microbial taxa responding differently to selected methods [28]. Considering rare members of the microbiome can contribute greatly to driving community dissimilarity, sample preservation methods should be chosen in accordance with targeted objectives [31-34]. As has been recently reviewed, there exist critical points that can affect the efficacy of methods for the preservation of soil microbiomes [35,36]. Besides storage temperature and duration as mentioned earlier, some of these include but not limited to choice of cryoprotectant and storage with soil matrix. A thorough assessment will help minimize any selective biases introduced to a particular taxon for a more comprehensive view of the microbial community and its functional potential. Cooperative interactions among national culture collections will accelerate technological development that further efficiently and comprehensively nation-led microbiome manage research projects.

## 5. Conclusion

The present review highlights the important role of microbial culture collections in the conservation of microbial resources for sustainable use. This article also sheds light on the need for extensive research toward the assessment of proper preservation methods and functional potential for microbiome samples. Taking inspiration from the networks of national culture collections in Japan and the United States expanding their scopes to accelerate technological development, collaboration among the domestic culture collections in the Republic of Korea is essential not only to conserve national microbial resources but also to support inclusive approaches to microbiome research and preservation. Microbial culture collection network within a country will lead to a country's industrial development beyond scientific impact.

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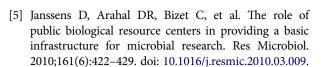
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