Available online at www.sciencedirect.com

ScienceDirect

Current Opinion in Virology

Phage banks as potential tools to rapidly and cost-effectively manage antimicrobial resistance in the developing world

Tobi Nagel[1], Lillian Musila[2], Milkah Muthoni[3], Mikeljon Nikolich[4], Jesca L Nakavuma[5] and Martha RJ Clokie[6]

Lower and middle-income countries seldom develop vaccines and therapeutics for their own populations and are dependent on supplies from industrialized countries, which are often hampered by financial or supply chain limitations. This has resulted in major delays in delivery with significant loss of life, as seen with the coronavirus pandemic. Since the vast majority of deaths from the antimicrobial resistance crisis are expected to occur in developing countries, there is an urgent need for in-country production of antibacterial therapies such as phages. Nationally controlled phage banks might provide such a solution since locally developed phage therapies tailored to endemic bacterial strains could offer cost-effective antibiotic alternatives.

Addresses
[1] Phages for Global Health, 383 62nd Street, Oakland, CA 94618, USA

[2] U.S. Army Medical Research Directorate-Africa, P.O. Box 606-00621, Nairobi, Kenya
[3] Kenya Medical Research Institute, P.O. Box 54840-00200, Nairobi, Kenya
[4] Wound Infections Department, Bacterial Diseases Branch, Walter Reed Army Institute of Research, 503 Robert Grant Avenue, Silver Spring, MD 20910, USA
[5] School of Biosecurity, Biotechnical and Laboratory Sciences, College of Veterinary Medicine, Animal Resources and Biosecurity, Makerere University, Kampala, Uganda
[6] Department of Genetics and Genome Biology, University of Leicester, University Road, LE1 7RH, UK

Corresponding author: Nagel, Tobi (tobi@phagesforglobalhealth.org)

Current Opinion in Virology 2022, **53**:101208

This review comes from a themed issue on **Bacteriophage therapy (2022)**

Edited by **Joana Azeredo** and **Jean-Paul Pirnay**

For complete overview about the section, refer Bacteriophage therapy (2022)

Available online 15th February 2022

https://doi.org/10.1016/j.coviro.2022.101208

Introduction

When global epidemics occur, developing countries receive essential medicines much later than industrialized nations do. For example, during the HIV/AIDS crisis, death rates in the US started to plummet a full decade before numbers in African countries began to abate because it took that long for the poorer countries to procure adequate supplies of antiretroviral drugs [1]. And while the US has more than enough coronavirus vaccines to fully vaccinate its entire population, African nations only have enough for an average of 5.8% of their populations [2,3]. A similar delay in drug delivery from industrialized countries will likely occur for the antimicrobial resistance (AMR) crisis, which by 2050 is expected to kill five times more people annually than either HIV/AIDS or COVID-19 have — with roughly 90% of the AMR deaths occurring in Africa and Asia [4••].

To avoid this catastrophe with AMR, developing countries must gain greater autonomy and capacity to develop and manufacture antibiotic alternatives for their own populations, rather than rely on receiving newly developed drugs from industrialized countries. This is a daunting task, but one that might be achieved by using naturally occurring bacteriophages (phages), which are viruses that can kill bacteria, including antibiotic-resistant strains. Collections of locally isolated, pre-characterized phages could be established as national or regional 'phage banks', which could be quickly deployed to address outbreaks of antibiotic-resistant infections. If such phage banks were managed by national health agencies with support from a network of local as well as international scientific institutions, they could provide rapid, cost-effective, in-country solutions for combating AMR. There is precedence for using phage banks in this way — the former Soviet Union did so for decades [5,6••]. Numerous public and private organizations in industrialized countries have already begun establishing phage banks to prepare for AMR [7,8•,9–11,12•,13,14]. It may be even more critical for developing nations to do so.

Phage banks are already used in Eastern Europe and beyond

Phages were used as antibacterial therapeutics even before antibiotics were discovered, and with the growing threat of AMR, interest in developing phage-based drugs has increased rapidly worldwide [15]. As natural viral predators of bacteria, phages work through different molecular mechanisms than antibiotics and thus are able to kill both antibiotic-resistant and antibiotic-sensitive bacteria. Phages also kill bacterial species more

specifically than broad-spectrum antibiotics do; as a result, phages can protect patients' microbiomes. Furthermore, unlike conventional antibiotics, phage products can be designed to minimize future bacterial resistance against them or even cause bacteria to become resensitized to antibiotics [16•].

Over the past century, a number of institutions have established large repositories of phages targeting a wide range of bacterial species. Perhaps the best known are the phage banks in the former Soviet Union, particularly at the Eliava Institute of Bacteriophages, Microbiology and Virology, now in the country of Georgia, which contains more than 1000 phages, and the Hirszfeld Institute of Immunology and Experimental Therapy in Poland, which has more than 850 phages [17,18]. Tens of thousands of patients have received phage treatment via these two institutions, often with individually customized phages selected for their ability to specifically kill each patient's infecting bacteria. In other cases, the broad collections of phages have been screened against disease-causing bacteria found to be circulating in the general population, then pre-set phage mixtures are made available as over-the-counter treatments. Importantly, those pre-set mixtures — typically called phage cocktails — are screened twice a year to make sure they are still active against the currently circulating bacteria; if not, the cocktails are updated [6••,19,20]. This approach of developing cocktails for the general population — rather than employing personalized phage therapy for individuals — might be particularly valuable for developing countries.

Other phage banks have been established largely to provide resources for laboratory research. The longest-standing of those include the Felix d'Herelle Reference Center for Bacterial Viruses in Canada (>400 phages), the American Type Culture Collection (ATCC) in the US (~350 phages), the German Collection of Microorganisms and Cell Cultures (also known as DSMZ; with ~450 phages), the National Collection of Type Cultures (NCTC) in the UK (>100 phages), and the Bacteriophage Bank of Korea (>1000 phages).

In the past few years, more banks have been set up specifically to provide phage therapy for patients, with funding primarily through governmental grants as well as some private donations. These include banks at the Queen Astrid Military Hospital in Belgium, both the U. S. Navy Medical Research Center — Biological Defense Research Directorate and the Walter Reed Army Institute of Research (with phages from each licensed to Adaptive Phage Therapeutics), the Hebrew University and Hadassah Medical Center in Israel, the Fagenbank in the Netherlands, Baylor College of Medicine in the US (called the Tailored Antibacterials and Innovative Laboratories for Phage [Φ] Research or TAILΦR initiative), and through the Australian Phage Biobanking Network [7,8•,9–11,12•,13,14]. These phage banks are typically set up to provide treatment for individual patients in situations when other treatment options have been exhausted. The phages can be approved through expanded access (often called 'compassionate use') regulatory pathways, which can apply to either emergency or non-emergency clinical cases. Some of these banks also use their phage collections to formulate phage cocktails for clinical trials and product development by more conventional regulatory pathways. In Belgium, the magistral phage pathway has been established, through which phage collections can be used for routine therapy for individuals, and this system is being considered by other countries in Europe.

A number of academic research groups and companies around the world have also established collections of phages, with some of those containing more phages than the formalized phage banks listed above and some banks focused on a narrow set of pathogens. A notable example is the SEA-PHAGES collection hosted at the University of Pittsburgh, through which more than 19 000 phages have been isolated, mostly targeting soil dwelling non-pathogenic mycobacteria, although including some phages that target related pathogenic mycobacteria that have been used for clinical phage therapy [21].

Phages are suitable for developing countries

Many aspects of phages make them particularly well-suited for use in the developing world. Most importantly, phages are able to kill antibiotic-resistant bacteria, a crucial treatment need as antibiotics fail at alarmingly high rates in developing countries. In addition, phages can be isolated locally using laboratory equipment that is readily available to scientists in developing countries. In fact, it is often best to isolate phages in the region where they will be used, since phages naturally adapt to target strains of bacteria that have evolved locally.

It is also relatively inexpensive to isolate, characterize and propagate phages. This aspect of phages could help overcome a key problem in the worldwide antibacterial drug market: conventional drug development is so expensive that companies cannot recoup their development costs by selling the antibiotics. In fact, several companies that have developed effective new antibiotics have recently gone bankrupt when sales revenues could not cover operating costs [22]. Antibacterial drugs simply cannot garner the same high prices as other drug classes. The problem is exacerbated by the fact that public health policies dictate that any new drugs that can successfully kill antibiotic-resistant bacteria should be held in reserve as last-line treatments — thereby minimizing any reimbursements that companies might be able to gain from developing good drugs. This has prompted global policymakers to propose new financial incentives for antibacterial drugs, with minimal success thus far.

Ultimately, local phage banks could enable national or regional control of phage-based drugs to help manage antibiotic-resistant outbreaks, thereby decreasing dependence on drug delivery from industrialized countries. Furthermore, if phages could be manufactured locally, this could decrease costs and logistics, since the phage products would not need to be shipped from overseas [23]. This might be accomplished through partnerships with existing organizations such as GAVI or GALVmed, which currently manufacture medicines for people as well as livestock in developing countries. Realistically, producing purified phages at scale would likely require collaborations with experts in developed countries, at least initially. But this technology transfer could be achieved over time, with the added benefit of providing new in-country jobs. Developing and manufacturing phage products locally could also increase community trust in the drugs, as long as safety and efficacy was demonstrated and the product quality was certified. An example of such "national pride" and associated social acceptance seems to be observed with a traditional (non-mRNA) COVID-19 vaccine that has recently been manufactured in Vietnam, Thailand and Brazil for clinical trials in those countries [24,25].

Phage banks should be national resources that follow international standards

Theoretically, the ideal organization for phage banks would be to have each bank managed by a central authority within each country. We envision that such banks would be hosted at government-funded universities or research institutions and supported with public and philanthropic funds, such as the examples currently being tested in Belgium, the US, Israel, the Netherlands and Australia. Phage banks within neighboring countries should be set up to coordinate rapid responses to regional outbreaks. The phages best suited to fight the outbreak should be used, regardless of the country where the phage originated — acknowledging that bacteria and phages can easily cross national boundaries. Thus, contracts and licensing agreements would need to be in place to recognize the phages as goods to be used for general public benefit, rather than as products controlled exclusively by private enterprise. Using in-country-derived phages in this way would be consistent with the Nagoya Protocol, an international agreement designed to 'provide for the fair and equitable sharing of benefits arising from the utilization of genetic resources' [26].

This would be a drastic shift from traditional drug development in which pharmaceutical companies drive the development of new antibacterial products. Instead, nationally appointed agencies would curate well-characterized endemic phages — and ideally provide phage products at cost or at supplemented rates. Realistically, public-private partnerships would likely be needed for some aspects of developing optimized phage cocktails, then manufacturing and disseminating them. The exact nature of such contractual details would undoubtedly be country-specific and complex, but establishing such agreements would be essential if phage banks are to be used to save lives in developing countries.

For phages to be safely delivered through such a centralized system, phage bank facilities would need standardized management systems following agreed-upon guidelines, such as those defined by the International Organization of Standardization (ISO), the Minimum Information About Biobank Data Sharing (MIABIS), or the International Society for Biological and Environmental Repositories (ISBER). These include requirements such as stable and secure databases, efficiently monitored storage facilities with access controls, as well as robust power supply backup systems. In addition, there would need to be standardized protocols for each step of the processing pipeline, including phage isolation, characterization, purification and routine quality control testing. Requirements for genomic and phenotypic data should also be standardized to allow easy phage selection, particularly in outbreak situations [27]. Ideally, well-characterized bacterial host strains would also be available, potentially secured through national AMR surveillance programs. Recently isolated bacteria could then be screened against the phage bank at regular intervals to select phage cocktails that could kill the problem bacteria — similar to the system that has been employed in the former Soviet Union for decades. All of this would require technically competent staffing, as well as oversight by governmental regulatory bodies, institutional review boards, and ethics committees relevant to each country.

Initial steps for phage banks in Africa are underway

Over the past 5–10 years, research groups in at least 13 countries in Africa have begun isolating phages, including in Eastern Africa (Ethiopia, Kenya, Tanzania, Uganda), Western Africa (Benin, Burkina Faso, Cote d'Ivoire, Ghana, Nigeria and Senegal), Northern Africa (Egypt and Tunisia), and Southern Africa (South Africa). In particular, scientists from institutions across Eastern Africa have already isolated more than 450 phages, though full characterization of most of those has yet to be completed (Nagel, personal communication).

With a critical mass of isolated phages in Eastern Africa, there has been interest from the local scientific community in establishing centralized phage banks there. While it might be ideal to eventually create an Eastern Africa regional phage bank, it seems most realistic to begin with country-specific phage banks, given that each nation has its own regulatory framework. National phage banks could be set up at institutions that already have the appropriate infrastructures and internationally recognized management systems in place. Some established

biorepositories might fit those criteria, as they adhere to rigorous regulations for storing isolated patient specimens. For example, in Nairobi both the Kenya Medical Research Institute (KEMRI) and the International Livestock Research Institute (ILRI) are ISO-certified institutions that maintain repositories of various biological samples, with KEMRI being the nationally mandated governmental research institute for human health and ILRI focusing on safe use of livestock and associated food products. In Uganda, the College of Health Sciences at Makerere University recently established a biorepository that handles specimens according to standard operating procedures dictated by ISBER. Biorepositories such as these might provide excellent settings for initial phage bank storage. Also, the close proximity between academics and the biorepositories would facilitate easier study of phages, allowing researchers to initiate projects to unravel specific aspects of phage biology.

To our knowledge, East African institutions involved in phage research have included:

- In Kenya — the Technical University of Kenya, the University of Nairobi, Jomo Kenyatta University of Agriculture and Technology, Kenyatta University, the Institute of Primate Research, KEMRI, ILRI, and the US Army Medical Research Directorate in Africa
- In Uganda — Makerere University, Kampala International University, Kyambogo University, Clarke University and Muni University
- In Tanzania — Kilimanjaro Christian Medical University College
- In Ethiopia — Jimma University

Many of the collected phages target bacteria causing infections in humans like antibiotic-resistant tuberculosis, pneumonia, and diarrhea — some of the leading causes of death in developing countries. Others kill bacteria carried by animals that can contaminate food products such as meat, fish, dairy or agriculture crops, which can subsequently cause illness in people. Thus, these phages could be utilized through a One Health approach, recognizing that bacteria can move between people, animals and the environment. Given that more antibiotics are used in livestock than in people worldwide, the use of phages in animals could further help with the AMR crisis by slowing the development of antibiotic resistance in food sources.

A paradigm shift is needed in AMR drug development to avoid huge death tolls

Past and current pandemics demonstrate that the developing world should not count on receiving needed medical supplies from abroad. The traditional strategy of developing drugs in the commercial sector in industrialized countries, then utilizing government support to provide those drugs to developing countries has not worked well to address public health needs with sufficient speed and efficacy. This will be an even bigger issue for antimicrobial drugs since, if nothing is done, the death toll from AMR infections will be manyfold larger than any previous infectious disease crises — including COVID-19. Instead, there must be a paradigm shift in how antibacterial drugs are developed and distributed to underserved populations. Phages may constitute much-needed antibiotic alternatives, and centrally organized phage banks may provide cost-effective, local mechanisms through which developing countries can effectively respond to antibiotic-resistant outbreaks.

Conflict of interest statement

Nothing declared.

Acknowledgements

Communications with numerous scientists worldwide helped inform the writing of this manuscript, including: Khaoma Barasa, Hamadi Boga, Benjamin Chan, Nina Chanishvili, Debora Charles, Sabrina Green, Abubakar Hoza, Ivan Ibanda, Deus Kamya, Anthony Kibera, Ruby Lin, Angela Makumi, Jean Pierre Mpatswenumugabo, Shawna McCallin, Sylvain Moineau, Arshnee Moodley, Nyambura Moremi, Ivy Mutai, Peter Muturi, Bashir Mwambi, Ritah Nakayinga, Janet Nale, Alice Namatovu, George Nasinyama, Jillani Ngalla, Atunga Nyachieo, Christine Rohde, Jessica Sacher, Frenk Smrekar, Nicholas Svitek, Marina Tediashvili, Denise Tremblay, and Jan Zheng.

This material has been reviewed by the Walter Reed Army Institute of Research. There is no objection to its presentation and/or publication. The opinions or assertions contained herein are the private views of the authors, and are not to be construed as official, or as reflecting true views of the Department of the Army or the Department of Defense.

This research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors.

References and recommended reading

Papers of particular interest, published within the period of review, have been highlighted as:

- • of special interest
- •• of outstanding interest

1. Nkengasong JN, Ndembi N, Tshangela A, Raji T: **COVID-19 vaccines: how to ensure Africa has access**. *Nature* 2020, **586**:197-199.

2. https://www.who.int/director-general/speeches/detail/who-director-general-s-opening-remarks-at-the-media-briefing-on-covid-19—7-october-2021.

3. https://ourworldindata.org/covid-vaccinations?country=.

4. •• *Tackling Drug-resistant Infections Globally: Final Report and Recommendations - The Review on Antimicrobial Resistance. Chaired by Jim O'Neill*. 2016

This is the foundational report commissioned by the UK government, which defined the expected global scope of the AMR crisis in terms of its death toll and economic impacts. The Global Action Plan on AMR as well as national AMR action plans worldwide have been developed largely based on information from this report.

5. Mie?dzybrodzki R, Hoyle N, Zhvaniya F, Łusiak-Szelachowska M, Weber-Da?browska B, Łobocka M, Borysowski J, Alavidze Z, Kutter E, Górski A, Gogokhia L: **Current updates from the long-standing phage research centers in Georgia, Poland, and Russia**. In *Bacteriophages: Biology, Technology, Therapy*. Edited

by Harper DR, Abedon ST, Burrowes BH, McConville ML. Cham: Springer International Publishing; 2018:1-31.

6. •• Abedon ST, Kuhl SJ, Blasdel BG, Kutter EM: **Phage treatment of human infections**. *Bacteriophage* 2011, **1**:66-85
This paper remains one of the most comprehensive reviews of phage therapy over the past century, including discussion of the early work carried out in Georgia, Poland, Russia, France and the US.

7. Djebara S, Maussen C, De Vos D, Merabishvili M, Damanet B, Pang KW, De Leenheer P, Strachinaru I, Soentjens P, Pirnay JP: **Processing phage therapy requests in a Brussels military hospital: lessons identified**. *Viruses* 2019, **11**:265 http://dx.doi.org/10.3390/v11030265.

8. • Schooley RT, Biswas B, Gill JJ, Hernandez-Morales A, Lancaster J, Lessor L, Barr JJ, Reed SL, Rohwer F, Benler S *et al.*: **Development and use of personalized bacteriophage-based therapeutic cocktails to treat a patient with a disseminated resistant *Acinetobacter baumannii* infection**. *Antimicrob Agents Chemother* 2017, **61**:e00954-17 http://dx.doi.org/10.1128/AAC.00954-17
This paper describes a high profile, complex case of a patient treated with phages in the US. It illustrates the value of phage banks, since the phages were obtained from three separate phage collections: from the military, a university, and a company. This demonstrated how different institutions can contribute rapidly and effectively to a successful phage therapy outcome.

9. Philipson CW, Voegtly LJ, Lueder MR, Long KA, Rice GK, Frey KG, Biswas B, Cer RZ, Hamilton T, Bishop-Lilly KA: **Characterizing phage genomes for therapeutic applications**. *Viruses* 2018, **10**:188 http://dx.doi.org/10.3390/v10040188.

10. Yerushalmy O, Khalifa L, Gold N, Rakov C, Alkalay-Oren S, Adler K, Ben-Porat S, Kraitman R, Gronovich N, Shulamit Ginat K *et al.*: **The Israeli Phage Bank (IPB)**. *Antibiotics* 2020, **9**:269 http://dx.doi.org/10.3390/antibiotics9050269.

11. Terwilliger AL, Gu Liu C, Green SI, Clark JR, Salazar KC, Hernandez Santos H, Heckmann ER, Trautner BW, Ramig RF, Maresso AW: **Tailored antibacterials and innovative laboratories for phage (phi) research: personalized infectious disease medicine for the most vulnerable at-risk patients**. *PHAGE* 2020, **1**:66-74 http://dx.doi.org/10.1089/phage.2020.0007.

12. • Lin RC, Sacher JC, Ceyssens PJ, Zheng J, Khalid A, Iredell JR: **Australian phage biobanking network: phage biobank: present challenges and future perspectives**. *Curr Opin Biotechnol* 2021, **68**:221-230 http://dx.doi.org/10.1016/j.copbio.2020.12.018
This paper describes the key factors that should be considered when utilizing a centralized phage bank, based on the initial experience of establishing a government-funded bank in Australia.

13. https://www.aphage.com/science/.

14. https://www.fagenbank.nl/english/.

15. Nikolich MP, Filippov AA: **Bacteriophage therapy: developments and directions**. *Antibiotics* 2020, **9**:135 http://dx.doi.org/10.3390/antibiotics9030135.

16. • Chan BK, Sistrom M, Wertz JE, Kortright KE, Narayan D, Turner PE: **Phage selection restores antibiotic sensitivity in MDR Pseudomonas aeruginosa**. *Sci Rep* 2016, **6**:26717 http://dx.doi.org/10.1038/srep26717
In this paper the authors demonstrate that directed biological pressure from phages can cause antibiotic-resistant bacteria to become re-sensitised to antibiotics. This opens up a new strategy by which phages may be used to extend the effective usage of existing antibiotics.

17. http://eliava-institute.org/phage-collection/?lang=en.

18. Żaczek M, Weber-Da?browska B, Mie?dzybrodzki R, Łusiak-Szelachowska M, Górski A: **Phage therapy in Poland - a centennial journey to the first ethically approved treatment facility in Europe**. *Front Microbiol* 2020, **11**:1056 http://dx.doi.org/10.3389/fmicb.2020.01056.

19. Kutter E, De Vos D, Gvasalia G, Alavidze Z, Gogokhia L, Kuhl S, Abedon ST: **Phage therapy in clinical practice: treatment of human infections**. *Curr Pharm Biotechnol* 2010, **11**:69-86 http://dx.doi.org/10.2174/138920110790725401.

20. Villarroel J, Larsen MV, Kilstrup M, Nielsen M: **Metagenomic analysis of therapeutic PYO phage cocktails from 1997 to 2014**. *Viruses* 2017, **9**:328 http://dx.doi.org/10.3390/v9110328.

21. https://www.hhmi.org/science-education/programs/science-education-alliance.

22. www.nytimes.com/2019/12/25/health/antibiotics-new-resistance.html.

23. https://www.gavi.org/vaccineswork/why-africa-needs-manufacture-its-own-vaccines.

24. https://www.nytimes.com/2021/04/05/health/hexapro-mclellan-vaccine.html.

25. https://www.news-medical.net/news/20210924/Inactivated-egg-based-Newcastle-disease-virus-vaccine-expressing-SARS-CoV-2-spike-protein.aspx.

26. https://www.cbd.int/abs/about/default.shtml/.

27. Millard Andrew: **Bacteriophage Introductions**. *PHAGE* 2020, **1**:37.