



Bacteriophage Therapy: a review and the Portuguese landscape

Terapia fágica: da evidência científica global à implementação em Portugal

Joana Alves^{1,2,3}, Ana Cláudia Carvalho¹, Carlos Capela^{4,5,6}, Betânia Faria^{2,7}, Rosalina Leite⁸, Luciana Meneses^{8,9}, Diana Priscila Pires^{8,9}, Joana Azeredo^{8,9}.

¹ Department of Infectious Diseases, Hospital de Braga, Unidade Local de Saúde de Braga, Braga, Portugal.

² Local Unit of the Program for Prevention and Control of Infection and Antimicrobial Resistance, Hospital de Braga, Unidade Local de Saúde de Braga, Braga, Portugal.

³ School of Medicine, University of Minho, Braga, Portugal.

⁴ Department of Internal Medicine, Hospital de Braga, Unidade Local de Saúde de Braga, Braga, Portugal.

⁵ Life and Health Sciences Research Institute (ICVS), School of Medicine, University of Minho, Braga, Portugal.

⁶ Clinical Academic Center, Braga, Portugal.

⁷ Pharmaceuticals Department, Hospital de Braga, Unidade Local de Saúde de Braga, Braga, Portugal.

⁸ CEB - Centre of Biological Engineering, University of Minho, Braga, Portugal.

⁹ LABBELS - Associate Laboratory, Braga, Portugal.

Autor correspondente: Joana Alves Email: joanamargaridaalves@gmail.com

DOI:10.65332/rpdi.v20.140 Recebido: 19 Jan 2026 Aceite: 16 Fev 2026 Publicado: 17 Mar 2026

ABSTRACT

This review provides a comprehensive overview of bacteriophage biology and classification, phage–bacteria interactions, and the mechanisms underlying phage–therapy efficacy, including its activity against biofilms and multidrug-resistant pathogens.

It further examines phage–antibiotic synergy, resistance dynamics, safety considerations, pharmacokinetic and pharmacodynamic aspects, and the role of phage banks.

Finally, the article discusses clinical evidence supporting phage therapy, emerging applications, and the Portuguese experience, highlighting regulatory, logistical, and clinical pathways that support the integration of phage therapy into modern infectious disease management.

Keywords: Bacteriophages; Phage Therapy; Antimicrobial Resistance; Chronic bacterial infections; Bacterial viruses.

RESUMO

Esta revisão oferece uma visão abrangente da biologia e classificação dos bacteriófagos, das interações fago-bactéria e dos mecanismos subjacentes à eficácia da terapia fágica, incluindo a atividade contra biofilmes e infeções por microrganismos multirresistentes.

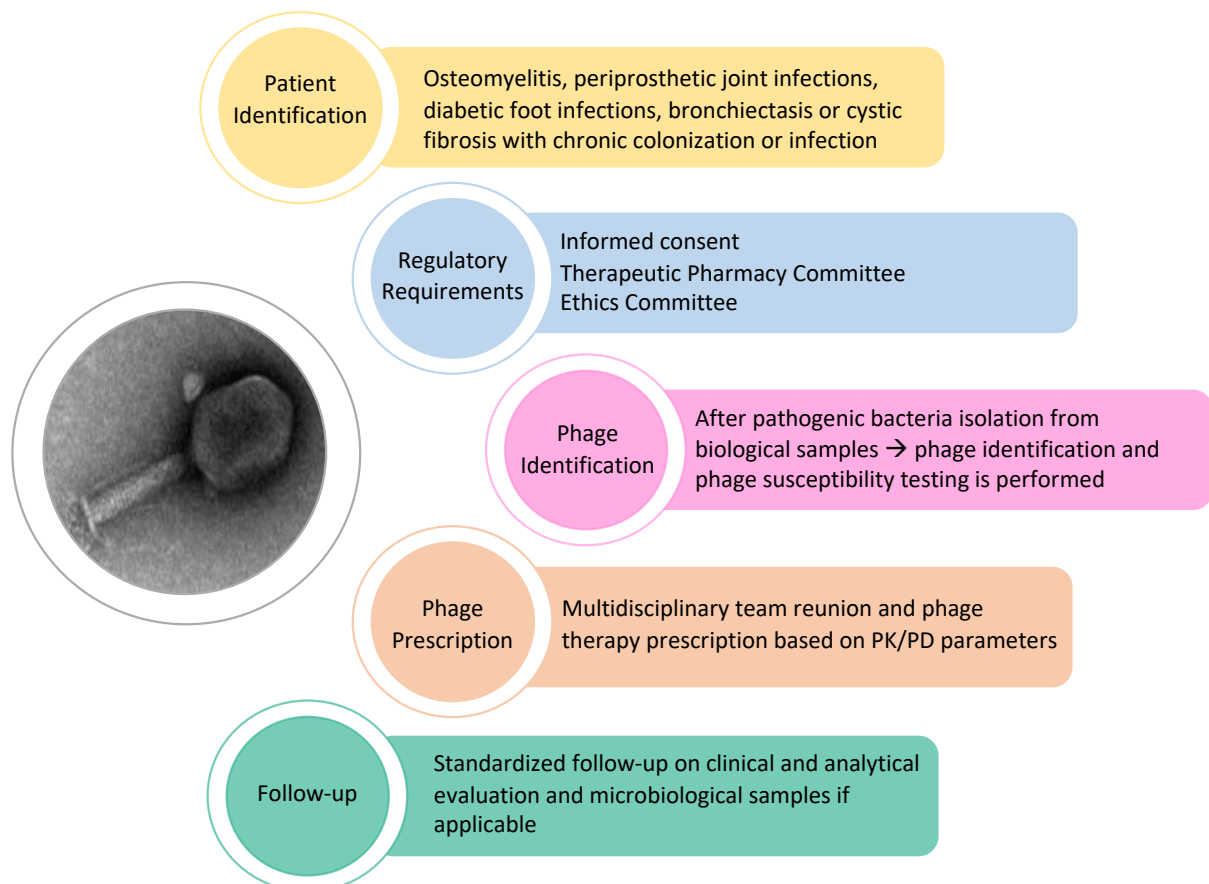
Descreve a sinergia fago-antibiótico, a dinâmica da emergência de resistências, as considerações de segurança, características farmacocinéticas e farmacodinâmicas e o papel dos bancos de fagos.

Por fim, o artigo discute as evidências clínicas que apoiam a eficácia da terapia com fagos, as aplicações emergentes e a experiência portuguesa, destacando os fluxos de trabalho regulatórios, logísticos e clínicos que apoiam a integração da terapia fágica na prática clínica.

Palavras-Chave: Bacteriófagos; Terapia fágica; Resistência antimicrobiana; Infeções bacterianas crónicas; Vírus bacterianos.

GRAPHICAL ABSTRACT

Phage therapy workflow developed by CEB in collaboration with the Queen Astrid Military Hospital and with Portuguese hospitals



Alves et al, Bacteriophage Therapy: a review and the Portuguese landscape, SPDIMC, 2026

Introduction

By the time of bacteriophages' discovery, in 1915 by Frederick Twort and in 1917 by Félix d' Hérelle^{1,2}, the research was focused on the treatment of bacterial infections, but the discovery and introduction of highly effective antibiotics in the following years largely displaced bacteriophage therapy from clinical practice. However, decades of extensive antibiotic use have selected for resistant microorganisms, and together with the limited development of new effective agents, this has become a major global health threat, with rising morbidity and mortality attributable to bacterial infections. In recent years, from 2018 to 2023, the incidence of resistant bacteria increased by more than 40.0%³. This scenario has led to a renewed interest in bacteriophages (phages) and their use in treating bacterial infections – phage therapy⁴.

Phage therapy represents a personalized therapeutic approach, with a growing number of successful clinical applications reported in recent years. Nevertheless, access to phage therapy remains limited by economic and regulatory barriers, as well as by geographic distance from specialized academic phage research centers.

Phage therapy has recently been approved in Portugal under a regulatory framework (INFARMED Deliberation n° 112/CD/2024) aligned with the Belgian model based on magistral preparations, representing a significant milestone for the clinical implementation of this personalized antimicrobial strategy. This development opens new opportunities to expand phage therapy as a complementary or alternative approach to antibiotics and to address the growing challenge of antimicrobial resistance, which is prevalent in Portugal⁵.

This review provides a comprehensive overview of phage biology and classification, phage–bacteria interactions, and the mechanisms underlying phage therapy efficacy, including its activity against biofilms and multidrug-resistant (MDR) pathogens. It further explores phage–antibiotic synergy, resistance dynamics, safety considera-

tions, pharmacokinetic and pharmacodynamic aspects, and the role of phage banks. Finally, we address the clinical evidence supporting phage therapy, its emerging applications, and the recent Portuguese experience, highlighting regulatory, logistical, and clinical workflows that support the integration of phage therapy into a modern infectious disease approach to infection.

1. Bacteriophage Principles

Bacteriophages, or phages, are viruses that infect bacteria and are considered the most abundant biological entities on Earth^{6–8}. They are also a natural and significant component of the human microbiome. Most known phages belong to the class *Caudoviricetes*, which comprises tailed viruses (figure 1) characterized by icosahedral capsids and double-stranded deoxyribonucleic acid (DNA) genomes⁹.

Previously, phages were traditionally classified based on their tail morphology into three major families: *Myoviridae*, characterized by long contractile tails; *Podoviridae*, possessing short non-contractile tails; and *Siphoviridae*, exhibiting long non-contractile tails¹⁰. However, the 2022 taxonomic update by the ICTV Bacterial Viruses Subcommittee has abolished these morphology-based families, which were replaced by genome-based taxonomy. Morphological descriptors such as “myovirus”, “siphovirus”, and “podovirus” are still used for descriptive purposes but no longer have formal taxonomic significance⁹.

This taxonomic revision resulted in extensive reorganization, including the establishment of one new order (*Crassvirales*), 22 new families, 30 subfamilies, 321 genera, and 862 species, reflecting a shift toward a genome-based classification system⁹.

Phages exhibit two primary life cycles, namely the lytic and lysogenic cycles. In both cases, phages start by attaching to bacterial surfaces via specialized tail proteins, enabling the injection of their genetic material^{11,12}. In the lytic cycle, phage DNA is transcribed and replicated within the host

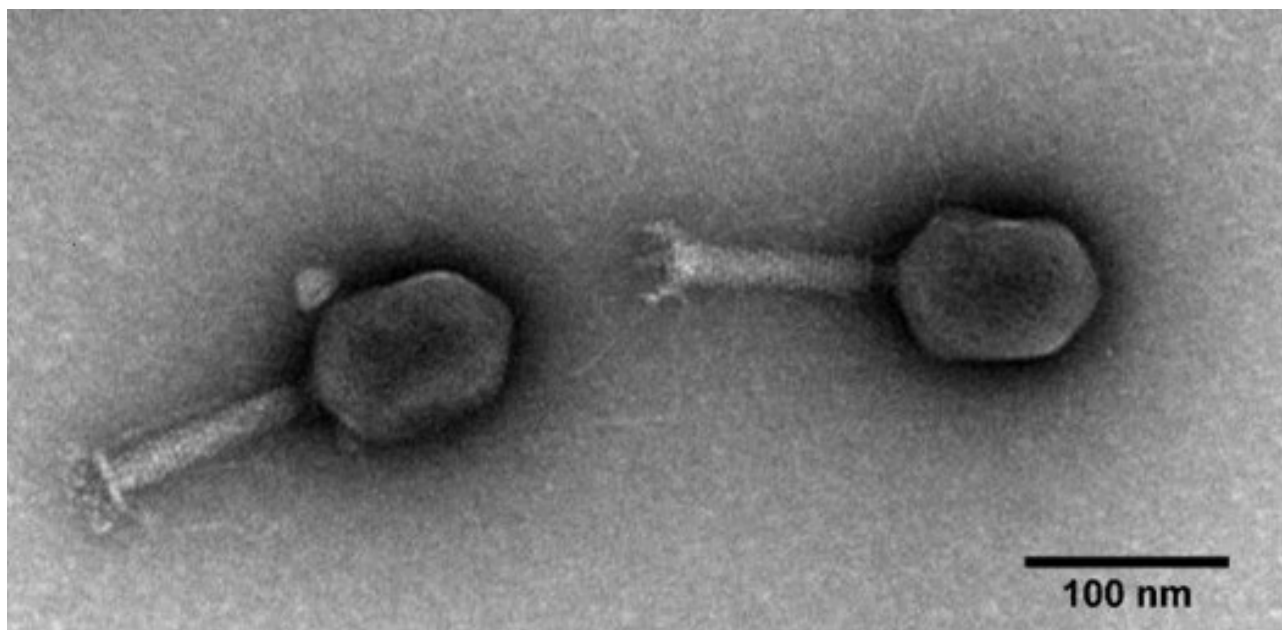


Figure 1. Transmission electron micrograph of an *Escherichia coli*-infecting bacteriophage with myovirus-like morphology from CEB's phage collection.

cell, leading to the assembly of new viral particles and, ultimately, bacterial lysis. The release of progeny phages enables subsequent rounds of bacterial infection^{11,12}. In contrast, in the lysogenic cycle, the injected phage genome integrates into the bacterial chromosome as a prophage and remains dormant, being passively replicated alongside the host DNA during normal bacterial cell division¹³.

Due to their capacity to specifically and exclusively infect and lyse bacterial cells, phages that exhibit a strictly lytic cycle (often called virulent phages) that do not transduce have attracted increasing attention across several fields, particularly regarding their potential application in phage therapy¹⁴. Phages that can alternate between a lytic and lysogenic cycle are called temperate phages and are avoided in phage therapy due to risks of bacterial persistence and gene transduction¹³.

Phages are known for their high degree of host specificity, targeting only a particular range of bacterial strains and replicating without affecting non-target cells. Consequently, phage-based approaches enable the elimination of pathogenic bacteria while preserving healthy commensal microbiota^{15,16}.

Bacterial resistance to phages frequently arises through the emergence of bacteriophage-insensitive mutants (BIMs). Although this resistance differs mechanistically from antibiotic resistance, bacteria can employ a wide range of antiphage defense systems, including receptor modification, superinfection exclusion, restriction-modification, and Clustered Regularly Interspaced Short Palindromic Repeats (CRISPR) mediated genome degradation, abortive infection systems, and antiphage signaling^{17–19}. Interestingly, while phage-resistant bacteria often arise rapidly under experimental conditions, the acquisition of resistance is commonly associated with significant fitness costs, frequently manifested as reduced growth rates, reduced virulence, and restored antibiotic sensitivity^{20,21}.

Moreover, phages have also evolved multiple counter-defense strategies to circumvent bacterial antiviral systems, including anti-CRISPR proteins, nucleotide modifications evading restriction-modification systems, and tail fiber mutations recognizing altered bacterial receptors^{19,22}. Consequently, co-evolutionary dynamics between phages and bacteria enable phage adaptation to BIMs, thereby restoring bacterial susceptibility and lytic activity despite initial resistance.

Phages can also act on complex environments, such as bacterial biofilms. Biofilms are clusters of bacteria encased in a self-produced matrix of extracellular polymeric substances (EPS). This protective mode of bacterial growth confers increased tolerance to antibiotics and host defenses, making biofilms more resistant than free-floating (planktonic) bacteria²³. Several phages have demonstrated the ability to inhibit biofilm formation and reduce pre-established biofilms²⁴. However, the biofilm EPS matrix itself can still provide substantial protection to bacterial cells, highlighting the need for combined or adjunctive treatment strategies^{25–27}.

2. Phage and antibiotic interactions

Phage therapy can be used alone or in combination with antibiotics, depending on which strategy offers the greatest therapeutic efficacy and the lowest likelihood of resistance development in each case^{28–30}.

It has been demonstrated that, in some cases, phages can be particularly effective against bacteria when combined with antibiotics. This phenomenon, known as phage–antibiotic synergy (PAS), is commonly observed *in vitro* as an increase in phage plaque size upon exposure of phage-infected bacteria to sublethal concentrations of antibiotics. Enlarged plaque size has been linked to enhanced phage adsorption rates, shortened latent periods, and increased burst sizes²⁸.

One of the mechanisms underlying PAS involves antibiotic-induced bacterial filamentation, a protective response of the bacteria when exposed to antibiotics and associated with inhibition of cell division. Antibiotics such as fluoroquinolones, β -lactams, and trimethoprim promote the formation of elongated, multinucleated bacterial cells, and these filamentous bacteria exhibit an increased surface area, which may enhance phage adsorption and subsequent infection³¹.

Filamentous bacteria may further enhance phage production either by delaying cell lysis—allowing

greater accumulation of phage particles before cell rupture—or by accelerating lysis, thereby increasing the rate of infection of neighboring cells³¹.

Although multiple PAS mechanisms have been described, no single mechanism fully explains the antibacterial efficacy observed in phage–antibiotic combinations. In addition, several studies have shown that phage–antibiotic combinations can potentiate antibiotic activity, reduce the emergence of resistance, or even restore the efficacy of antibiotics against resistant bacterial strains³².

By simultaneously targeting distinct bacterial structures, phages and antibiotics impose multiple selective pressures, which may explain the reduced likelihood of emergence of resistance. This principle is analogous to that underlying phage cocktails, where multiple phages are selected and used simultaneously to target different bacterial receptors, reducing the chance of resistance emergence^{33,34}.

Additional synergistic mechanisms include enhanced antibiotic penetration into biofilms, as some phages encode enzymes that can degrade the biofilm matrix, thereby facilitating antibiotic access to embedded bacterial cells³⁵.

Despite these synergistic effects, antagonistic interactions between phages and antibiotics have also been reported. Because virulent phages depend on host cellular machinery involved in transcription, translation, and DNA replication, antibiotics that inhibit these processes may impair phage replication, resulting in phage–antibiotic antagonism^{36–38}.

Furthermore, certain antibiotics may indirectly reduce phage efficacy by enhancing bacterial defense systems^{31,39}. For example, some antibiotics modulate CRISPR–Cas immunity, which provides adaptive protection against phage infection through spacer acquisition and targeted cleavage of phage DNA^{31,40}.

Bacteriostatic antibiotics have been shown to enhance CRISPR-mediated immunity by prolonging the eclipse period, reducing phage production,

and extending the temporal window for spacer acquisition^{31,39}.

Given the complexity and variability of phage–antibiotic interactions, *in vitro* evaluation is essential before clinical application. Commonly employed assays include time–kill curves and phage–antibiotic interaction analyses, such as synograms. As demonstrated by Liu et al., PAS is strongly influenced by multiple factors, including the antibiotic mechanism of action, stoichiometry, resistance determinants, phage-specific parameters (e.g., burst size), and environmental conditions⁴¹.

3. Tackling the challenges of antibiotic resistance

Antimicrobial resistance is a global public health issue and threatens the effective treatment of bacterial infections³.

A key concern is the rising number of infections caused by invasive Gram-negative bacilli, which are linked to increased mortality rates⁴². The World Health Organization has updated its list of bacterial priority pathogens, highlighting the urgent need for investment, especially in treating infections caused by *Acinetobacter baumannii* resistant to carbapenems, *Enterobacteriales* resistant to third-generation cephalosporins, and other carbapenem-resistant strains⁴³.

Phage therapy narrows the gap between MDR infections and available treatment options. Various centers offer patients phage therapy for multiple bacterial infections, including those caused by *Acinetobacter baumannii*, *Burkholderia cepacia*, *Citrobacter spp.*, *Enterobacter aerogenes*, *Enterococcus faecalis*, *Enterococcus faecium*, *Escherichia coli*, *Klebsiella pneumoniae*, *Morganella spp.*, *Proteus mirabilis*, *Proteus vulgaris*, *Pseudomonas aeruginosa*, *Salmonella spp.*, *Serratia marcescens*, *Shigella spp.*, *Staphylococcus aureus*, *Staphylococcus epidermidis*, *Staphylococcus saprophyticus*, *Streptococcus pyogenes*, *Streptococcus salivarius*, *Streptococcus sanguis*, and *Stenotrophomonas maltophilia*, among others⁴⁴.

In various environments like oceans, soil, freshwater, the human body, fermented foods, and waste, there are countless phages, each capable of infecting specific bacterial strains. Compared to antibiotics, phages are widely distributed throughout the biosphere and exhibit high diversity. However, each phage typically infects only a narrow range of bacterial strains. Because bacterial populations show substantial variation even within the same species, effective phage therapy usually requires a combination of different phages in a so-called phage cocktail. This highlights the inherently personalized nature of phage therapy, in which efficacy depends on the specific interaction between the phage receptor-binding machinery and the surface receptors expressed by the patient's bacterial isolate. In many clinically important pathogens, these receptors vary substantially between strains (even within the same species), so a phage (or a cocktail) that is effective in one patient may fail in another, despite infection by the same bacterial species^{45,46}. This issue is particularly pronounced in capsulated bacteria, where the capsular polysaccharide (CPS) often acts as a dominant adsorption determinant and shows extensive strain-to-strain diversity. In *Klebsiella pneumoniae*, capsule (serotype/K-locus) differences can strongly affect phage susceptibility⁴⁷. Similarly, in *Acinetobacter baumannii*, a critical-priority MDR pathogen, the capsule is frequently a primary receptor for phage attachment, and CPS structures are highly diverse (with large numbers of distinct capsule loci/types described), meaning that most phages infect only a limited subset of clinical isolates unless cocktails are tailored to the infecting strain's capsule type⁴⁸.

4. Phage Banks

The renewed interest in phage therapy has led to increased attention toward historical phage banks, alongside the emergence of new laboratories dedicated to phage research. Consequently, there has been a rapid expansion in the number of phage collections, ranging from small laboratory-based repositories to large institutional banks. However,

this growth has occurred with relatively limited coordination and a lack of standardized cataloguing practices, and to date, no comprehensive international registry of phage collections exists⁴⁹. Enhanced coordination and data sharing among phage researchers are therefore critical to expanding access and increase the likelihood of successful therapeutic matching in phage therapy. Many existing collections operate as service-providing culture collections or Biological Resource Centers (BRCs), acting as custodians of phage diversity, supplying phages for a fee, and supporting both internal and external research activities⁴⁹.

Among the most well-known phage banks are those originating from the former Soviet Union, particularly the Eliava Institute of Bacteriophages, Microbiology and Virology in Georgia, dating back to 1923, which maintains over 1,000 phages, and the Hirszfeld Institute of Immunology and Experimental Therapy in Poland, established in 1952, which holds more than 850 phages^{50,51}.

Additional phage banks have been established over time primarily to support laboratory research, including the Félix d' Hérelle Reference Center for Bacterial Viruses in Canada (>400 phages), the American Type Culture Collection (ATCC) in the United States (approximately 350 phages), the German Collection of Microorganisms and Cell Cultures (DSMZ; approximately 450 phages), the National Collection of Type Cultures (NCTC) in the United Kingdom (>100 phages), and the Korean Phage Bank (>1,000 phages)⁵⁰.

Although efforts have been made to centralize phage collections, the establishment of national or international infrastructures remains challenging. As phage therapy has not yet been fully integrated into routine clinical practice, securing sustained public or private investment for centralized repositories is difficult⁴⁹.

Also, a critical requirement for the long-term viability of phage biobanks is the availability of reliable cryopreservation strategies. Despite their importance, standardized and universally ac-

cepted protocols for phage cryopreservation are still lacking^{52,53}.

5. Phage Therapy Efficacy

Phage therapy has a wide range of applications alone or in combination with antibiotics, in different anatomic sites: urinary tract infections; genital tract infections; postsurgical infections; respiratory tract infections; soft and skin tissue infections; bone and joint infections; cardiovascular and intravascular infections⁵¹. Phages can also penetrate complex bacterial communities, such as biofilms, and infect persistent or hard-to-reach cells (e.g., areas with poor blood circulation)²⁴. In addition, phages can respond effectively to the emergence of resistant bacterial mutants, since phages replicate in situ and, under selective pressure, spontaneous mutations within the phage population can give rise to variants with advantageous traits, such as expanded host range or enhanced infectivity^{4,54,55}.

The inappropriate clinical use of phages between 1920 and 1930 raised important concerns about safety and efficacy²⁹, contradicted by recent research^{56,57}, where the efficacy of phage therapy against MDR bacteria has been demonstrated by several in vitro studies, showing significant reduction in bacterial loads and biofilms after phage treatment.

There is also evidence of the safety and efficacy of phages in reducing bacterial loads in in vivo models of infections, such as lung infections and wound infections^{62–66}. Besides the promising results from in vitro and in vivo studies, there is an increasing number of case studies reporting encouraging results of patients with life-threatening antibiotic-resistant bacterial infections that have been successfully treated with phage therapy^{63–65}.

Table I (see supplementary material at the end of the article) summarizes reported clinical cases and case series describing the use of phage therapy across multiple infection sites to date.

Osteoarticular infections represent the largest body of published clinical experience with phage

therapy. In this setting, phage therapy is predominantly administered locally, as summarized in Table I, enabling high local phage concentrations despite the absence of standardized pharmacokinetic data. The heterogeneous phage dosing parameters in clinical literature reflect the lack of standardized guidelines and the individualized nature of its use.

Osteoarticular infections are difficult to treat, as microorganisms adhere to devitalized bone and prosthetic material, forming mature biofilms^{108,122–124} with limited antibiotic penetration and high bacterial persistence. Phage therapy may target bacteria embedded within biofilm partly through the production of depolymerases that degrade the biofilm matrix and through local phage amplification at the site of infection²⁴.

Cardiovascular and intravascular infections are rare but severe, including endocarditis, prosthetic valve infections, and vascular graft infections. In this context Phage therapy has been almost exclusively used as salvage adjunctive therapy (commonly intravenous), typically combined with prolonged systemic antibiotics and, when feasible, surgery. The observed tolerability of intravenous phage administration, even in critically ill patients, is a clinically relevant finding^{89–98}.

Clinical experience with phage therapy in respiratory infections remains limited but has increased in recent years. Phage therapy is predominantly delivered by nebulization, and doses are rarely reported, as summarized in Table I. In this setting, even when phage therapy could not achieve complete microbiological eradication, the majority of patients showed consistently clinical benefit^{99,101,102,105–107}.

The lung represents a dynamic compartment with rapid clearance mechanisms¹²⁵, making pharmacokinetics particularly relevant yet poorly characterized as reflected by inconsistent dosing and limited pharmacokinetic data reported in published clinical cases.

In urinary tract infections, different administration routes are reported, such as oral, rectal, and intravesical instillations. Again, data on specific dosing schedules is rarely reported. Of notice, in a notable proportion of cases, phages were administered without concomitant antibiotics^{112–115,121}.

In summary, Table I illustrates an important wide range of concomitant antimicrobial agents spanning from narrow- to broad-spectrum antibiotics. No evidence of phage-antibiotic antagonism was reported.

Across the reported cases, phage selection was consistently guided by phagogram testing, and systemic antibiotics are almost always prescribed concomitantly with phage therapy. There was marked heterogeneity in phage dosing, routes of administration, and treatment duration. These findings reflect a prescription experience-based and highlight the need of standardized treatment protocols. As discussed by Abedon et al.¹²⁶, phages are self-amplifying biological agents whose replication depends on the local bacterial burden, suggesting that precise dosing may be less critical than for conventional antimicrobials and that repeated administration at the site of infection may not always be necessary.

Several case reports document the use of cocktails comprising two or more phages^{72,74,75,77,82,84,87,95,96,106,107} targeting the same bacterial strain and administered either simultaneously or sequentially to avoid the likelihood of resistance emergence during phage therapy¹²⁷. Importantly, even when phage resistance is documented, it does not necessarily correlate with clinical failure, as demonstrated by Yang et al. and Li et al.^{99,105}.

Furthermore, across reported clinical cases, phage therapy was associated with consistently low rates of phage-related adverse events, and no dose-dependent toxicity has been described to date^{127,128}.

Personalized phage therapy, while offering high specificity and the potential for targeted therapy,

is inherently time-consuming. The process typically requires bacterial isolation, phage susceptibility testing, regulatory approval, and often customized phage preparation. These steps may take several days to weeks, limiting its feasibility in acutely septic patients in need of immediate therapy^{130–132}. This limitation is particularly relevant in cardiovascular and pulmonary infections, where patients frequently present with acute or rapidly progressive disease. Consistent with this constraint, the summarized cases in Table I predominantly describe phage therapy as adjunctive therapy, initiated after prolonged antibiotic exposure, infection recurrences, or when surgical options were limited.

Interestingly, phage therapy use is not restricted to infection treatment, and it can play an important role as a prophylactic agent or microbiome regulator in colonized patients, as well as in the construction of antivenoms and biocontrol in food manufacturing, among other applications^{44,133–135}.

6. Phage Pharmacokinetics and Pharmacodynamics

Therapeutic optimization of phages is essential to achieve the desired therapeutic effect while minimizing the occurrence of toxicity and resistance¹³⁶. In this context, a thorough understanding of their pharmacokinetic (PK) and pharmacodynamic (PD) properties is required. Pharmacokinetics refers to the mechanisms involved in determining phage concentrations within the organism, whereas pharmacodynamics describes the effects that phages may exert on the human body¹³⁷.

The wide diversity of phages, formulations, and treatment regimens hinders the establishment of general conclusions regarding their *in vivo* pharmacokinetic/pharmacodynamic (PK/PD) characteristics and bioavailability¹³⁶. Despite the extensive use of phages in several countries, the lack of PK/PD information remains a persistent challenge for phage therapy^{136,138,139}. As biological entities capable of rapid self-replication at the site of infection and characterized by single-hit bacte-

rial killing kinetics, phages exhibit highly complex PK/PD behavior^{136,139,140}.

The main pharmacokinetic and pharmacodynamic elements to be considered in phage therapy include their pharmacological characteristics, the route of administration, the site of bacterial infection, the relative concentrations of phages and bacteria at different anatomical locations (including the site of infection, when feasible), and the interaction with the host immune system¹³⁷.

6.1 Phage Pharmacokinetics

In comparison with antibiotics, phages are living biological entities that replicate in the presence of susceptible bacteria, exhibiting PK profiles that are considerably more complex than those of conventional antibacterial agents¹³⁹. Therefore, sensitive and validated quantification methods are required for appropriate PK/PD monitoring of phages¹³⁶.

To describe PK in phage therapy, it is necessary to consider how phages interact with the host organism, including the evaluation of absorption, distribution, metabolism, and elimination processes, as well as phage dosing strategies¹³⁷.

The phage dose is the number of particles of each administered phage solution and forms the basis for PK determination¹³⁶.

Several techniques have been employed to investigate the *in vivo* PK properties of phages, including the measurement of phage titers in body fluids, as well as phage labeling as an alternative approach to assess their distribution¹³⁶.

Intravenous administration of phages has proven to be effective, providing good tissue penetration, particularly in tissues affected by inflammatory processes, with a consequent increase in endothelial barrier permeability¹³⁷.

Bacteriophages can circulate and accumulate in different organs and tissues¹³⁶. They have been detected in blood, bronchoalveolar lavage fluid, feces, lungs, heart, liver, kidneys, spleen, and brain

tissue, indicating that they are capable of crossing the blood–brain barrier¹³⁶. Although phages appear to access multiple tissues with relative ease, it remains unclear whether therapeutic concentrations at sites of infection can be achieved regardless of the administration route¹³⁶.

Phages accumulate in the liver and spleen, which are the organs responsible for their inactivation and elimination¹³⁷. The elimination of phages through urine is low¹³⁷.

It is important to note that the innate immune system also plays a fundamental role in phage elimination, mainly through phagocytosis¹⁴⁰.

6.2 Phage Pharmacodynamics

PD focuses on defining the relationship between phage concentration and bacterial elimination and on evaluating potential adverse effects associated with phage therapy. Primary PD studies aim to determine the effective phage concentration capable of reducing or eradicating bacterial loads to restore health, commonly quantified using the multiplicity of infection, which represents the ratio of phages to bacterial cells¹³⁶. Because not all administered phages successfully interact with bacteria, PD modelling must account for adsorption rates and bacterial density, reflecting the dynamic predator–prey relationship unique to phage therapy. Two main therapeutic strategies are described: passive therapy, which relies on administering phages in excess of the bacterial burden, and active therapy, which leverages phage replication at the infection site once bacterial and phage threshold concentrations are exceeded¹³⁷. Secondary PD studies address safety considerations, including toxicity linked to endotoxin contamination and broader effects on the microbiota, immune system, and eukaryotic cells, underscoring the need for rigorous purification and evaluation¹³⁷. Together, these PD principles highlight that, unlike antibiotics, phage exposure and response are interdependent, making careful optimization of phage dosing and bacterial targeting essential for effective and safe clinical application¹³⁷.

7. Safety profile and Adverse Effects

Phage therapy has demonstrated safety, tolerability, and efficacy in multiple clinical trials targeting different bacterial infections, resulting from its specific targeting of bacteria without affecting mammalian cells¹²⁷. Even at high concentrations, phage therapy has demonstrated excellent biosafety profiles¹²⁸.

Although clinical trials suggest that phage therapy is generally safe and well tolerated across a range of diseases and patient profiles, it is essential to recognize that patient-specific characteristics—such as immune status and comorbidities—as well as the phages used, the dosing regimen, and the route and/or method of administration, may influence the occurrence and severity of adverse reactions¹²⁷.

In cases where adverse effects were reported—such as pulmonary exacerbations in cystic fibrosis, bruising, dizziness, fatigue, headaches, weight gain, cough, nasal congestion, postoperative wound complications, and gastrointestinal disturbances—they were often nonspecific and their direct relation to phage treatment remains unproven¹²⁷. The adverse effects may have been attributable to other factors, such as the underlying clinical condition and/or concomitant treatments, rather than to phage therapy^{71–140}.

One of the main safety concerns regarding phages involves the presence of residual endotoxins in preparations¹²⁸. Endotoxins are lipopolysaccharides found in the outer membrane of Gram-negative bacteria and are released during bacterial lysis or death¹²⁸. These endotoxins are highly immunogenic and can raise body temperature, reduce white blood cell counts, and even cause shock¹²⁸. Therefore, effective endotoxin removal processes are necessary to meet stringent safety standards and facilitate the standardized use of phage therapies¹²⁸.

Another challenge lies in the production of anti-phage antibodies, which can limit the therapeutic efficacy of phage preparations¹²⁸. In mammalian

hosts, serum immunoglobulin M and immunoglobulin G antibodies can reduce or inhibit phage activity, respectively¹²⁸. The host immune response to bacterial infection may lead to rapid clearance of phages from the system, thereby compromising their therapeutic efficacy¹²⁸.

Although phages have been reported to induce only mild immune responses, further studies are still required¹²⁸.

Finally, phages are composed of proteins and nucleic acids, which can interact with the human immune system and elicit immunogenicity and potentially trigger allergic reactions¹²⁸.

Pharmacovigilance efforts, including post-marketing surveillance and adverse event reporting systems, are essential for monitoring the safety of phage-based products in real-world settings and for identifying any rare or unexpected adverse effects that may arise over time¹²⁷.

8. Local Experience - Phage therapy in Portugal

Phage therapy is already a commonly used practice in the treatment of antibiotic-resistant infections in Australia, Belgium, France, Georgia, Germany, Poland, Russia, and the United States of America in dedicated phage therapy centers^{141–142}.

A framework allowing the use of phage therapy in clinical practice was recently approved in Portugal (INFARMED Deliberation n^o 112/CD/2024). According to this resolution, phage therapy may be used as a magistral preparation for individual patients, in accordance with Article 3(1) of Directive 2001/83/EC (magistral formula) and Article 5 (direct responsibility of a healthcare professional). Only non-genetically modified bacteriophages are covered under this framework¹⁴³.

The magistral preparation must be carried out exclusively in a hospital pharmacy, based on an individualized medical prescription for a specific patient, and in compliance with Good Practices for

the Preparation of Manipulated Medicines. The entity responsible for producing the active agents (bacteriophages) must provide well-characterized bacteriophages, maintain a master phage bank, and perform quality assessments at two levels: genetic control and quality parameter control of the different production batches, ensuring the maintenance of appropriate batch records and certificates of analysis¹⁴³.

Once the clinical team decides to propose phage therapy, its use requires validation by the Ethics Committee, the Pharmacy and Therapeutics Committee, and the local Program for the Prevention and Control of Infections and Antimicrobial Resistance unit. Following each treatment, the outcome must be reported to INFARMED¹⁴³.

In Portugal, several laboratories and companies are dedicated to phage research and production. The Centre of Biological Engineering (CEB) at the University of Minho is a national reference in phage research with a substantial body of published literature, including review articles and pre-clinical studies addressing both the efficacy and the challenges of phage therapy^{25,26,145,146}. CEB is also involved in personalized phage production for Portuguese patients, in collaboration with the Queen Astrid Military Hospital - QAMH (Belgium) and, in 2025, formally announced the creation of a laboratory dedicated to the production of therapeutic phages¹⁴⁴.

Technophage, a Lisbon-based biotechnology company, has nearly two decades of experience in the development and manufacture of phage cocktails. The company has already supported compassionate-use treatments in countries such as France and Israel¹⁴⁷.

LxBio is another Portuguese biopharmaceutical company engaged in phage innovation and integrated into international networks such as PhageEU, although it does not yet have a publicly disclosed clinical pipeline¹⁴⁸.

In addition, numerous national academic laboratories generate knowledge, tools, and biomaterials

that may support phage therapy programs. The PhaBRIC laboratory at the Institute for Medicines Research (iMed), University of Lisbon, focuses on phage biology and lytic proteins, studying mechanisms of bacterial lysis and phage–host interactions, and developing phage-derived agents targeting *Enterococcus spp.*, *Mycobacterium tuberculosis* and *Staphylococcus spp.*¹⁴⁹.

The BeSurf group at i3S, University of Porto, specializes in BioEngineered Surfaces, developing biomaterials and surfaces functionalized with phages or phage-derived elements to prevent device-associated infections and osteomyelitis, exploring targeted and antibiotic-free approaches¹⁵⁰.

Finally, the International Iberian Nanotechnology Laboratory (INL), located in Braga, develops projects such as “Phages on Chip”, which integrate bacteriophages into microfluidic devices for diagnostic applications and for addressing the antimicrobial resistance crisis¹⁵¹.

To date, CEB has contributed to more than 20 phage therapy treatments in Portugal targeting strains of *Mycobacterium abscessus*, *Pseudomonas aeruginosa* and *Staphylococcus aureus*.

In the remainder of this section, we describe the workflow implemented at CEB that goes from the identification of a suitable patient to the follow-up of those receiving phage therapy.

Box 1: Phage therapy workflow developed by CEB in collaboration with the Queen Astrid Military Hospital and with Portuguese hospitals.

Identification of Suitable Patient

Patient identification is conducted in both inpatient and outpatient units during routine medical appointments. Following initial selection, an Infectious Diseases specialist evaluates the patient’s clinical condition and discusses the case within a multidisciplinary team, according to the patient’s condition and the source of infection.

Eligible candidates are then referred to the CEB via email. The physician responsible for the case must complete the patient’s clinical file and submit it to CEB. This file is subsequently reviewed by a multidisciplinary evaluation team, composed of members from both CEB and the QAMH. The team determines whether the clinical condition justifies the use of phage therapy, considering patient characteristics, infection site, microbiological data, and other relevant factors.

In general, phage therapy is considered for patients with chronic osteomyelitis, periprosthetic joint infections without curative surgical options, diabetic foot infections caused by difficult-to-treat bacteria, and patients with structural lung diseases (such as bronchiectasis or cystic fibrosis) presenting chronic colonization or infection with no clinical improvement under standard-of-care treatments.

Regulatory Requirements

If treatment is approved and active phages against the patient’s strain are available, the patient is provided with an informed consent form. The treating physician must then submit the required documentation for review by the hospital’s Therapeutic Pharmacy Committee and the local Ethics Committee.

Clinical Procedures

Following approval by the Therapeutic Pharmacy Committee and the Ethics Committee, collection of bacteriological samples from the infection site is scheduled. Pathogenic bacteria are isolated from these samples and forwarded to CEB, where phage susceptibility testing is performed on all recovered bacterial strains.

When multiple phages demonstrate activity against the same strain or strains, those with the highest efficiency of plating (EOP) are prioritized. If uncertainty remains regarding optimal phage selection, additional pharmacokinetic and efficacy assays are conducted. These assays evaluate both individual phages and phage combinations to identify the formulation that most effectively reduces bacterial burden.

The selected phages are produced and supplied by the QAMH. Upon receipt, phage stocks are stored at 2–8 °C to preserve viability. Final preparation is performed according to the medical prescription and maintained at 2–8 °C until administration, for a maximum period of one week.

Medical Prescription

Phage therapy is prescribed by the treating physician, with the dose, administration frequency, and treatment duration being previously discussed within a multidisciplinary team involving an Infectious Diseases Specialist and agreed upon with the QAMH team.

Follow-up

After completion of treatment, the physician responsible for the case completes a standardized follow-up form including information from the pre-treatment, treatment, and post-treatment periods, considering microbiological analyses, clinical symptoms, concomitant medications, and any potential adverse events related to therapy. The physician also reports whether, following phage therapy, the patient eradicated the target strain, experienced clinical improvement, or showed no noticeable changes during treatment.

Following treatment, additional experimental assays may be conducted at CEB using serum or sputum samples collected from the patient before, during, and after therapy. Serum-based assays are used to determine whether anti-phage antibodies were generated during treatment and, when present, to quantify antibody titers.

In patients with respiratory tract infections, sputum-based assays are performed to evaluate the reduction or eradication of the target bacterial strain and to assess potential changes in the strain's antibiotic susceptibility over the course of therapy.

In both cases, phage susceptibility testing is repeated using the phages administered during treatment to determine whether any alterations in phage sensitivity occurred during therapy.

Conclusion

This review provides a comprehensive overview of phage therapy, spanning fundamental phage biology, pharmacokinetic and pharmacodynamic considerations, and available clinical evidence regarding efficacy and safety.

There is marked heterogeneity in phage dosing, routes of administration, and treatment duration, reflecting the current reliance on experience-based prescribing and highlighting the need for standardized treatment protocols.

Overall, the majority of reported cases described clinical resolution, with consistently low rates of phage-related adverse events, supporting the favorable safety profile of phage therapy.

Finally, in Portugal, several laboratories and companies are actively engaged in phage research and production, and a growing body of clinical experience is emerging. Together, these developments support the progressive integration of phage ther-

apy into modern infectious disease management frameworks.

Author Contributions

Joana Alves and Joana Azeredo: Conceptualization, writing—original draft preparation, writing—review and editing.

Ana Cláudia Carvalho: Conceptualization, writing—review and editing.

Carlos Capela, Betânia Faria, Rosalina Leite, Luciana Meneses, and Diana Priscila Pires: Writing—original draft preparation, writing—review and editing.

All authors have read and agreed to the published version of the manuscript.

Funding / Sponsorship

The authors received no financial support for the research, authorship, and/or publication of this article.

Ethics statement

This study is a review of previously published literature and did not involve human participants or animals. Therefore, ethical approval and informed consent were not required.

Conflicts of Interest

The authors declare no conflict of interest.

Table I. Clinical use of bacteriophage therapy: reported cases and case series

| Author, year | Infection | <i>n</i> | Microbiology | Phage preparation ¹ | Route of administration, dosage, frequency and duration | Combined antibiotics | Reported outcome | Adverse events | Ref. |
|----------------------------------|----------------------------|----------|---|--------------------------------|--|--|--|---|------|
| Osteoarticular infections | | | | | | | | | |
| Eschweiler et al., 2025 | Prosthetic joint infection | 3 | MDR Gram - <i>S. epidermidis</i> , <i>S. capitis</i> , <i>Cutibacterium acnes</i> | Commercial phage cocktail | Oral and intralesional 10–30 mL per dose (variable) Once or twice daily 1–8 days (short patient-dependent courses) | Ampicillin/sulbactam Teicoplanin Amoxicillin/clavulanate | Resolution | Not reported | [66] |
| Cammuso et al., 2025 | Prosthetic joint infection | 1 | <i>S. epidermidis</i> | Personalized phage therapy | Intra-articularly and intravenously 7×10^9 PFU/dose, IA, twice daily, 7 days and IV, daily 14 days | Daptomycin Rifampin Linezolid | Resolution under suppressive antibiotic therapy | Hypotension, hypertension, low-grade fever, chest pain, rigors, and wheezing; increased liver enzymes | [67] |
| Wahl et al., 2025 | Prosthetic joint infection | 1 | MRSA | Personalized phage therapy | Intra-articularly and intravenously 1×10^7 PFU/mL, 10 days | Daptomycin Trimethoprim/sulfamethoxazole | Resolution; death at 6 months unrelated to infection | Not reported | [68] |
| Doub et al., 2023 | Prosthetic joint infection | 1 | <i>Enterococcus faecalis</i> | Personalized phage therapy | Intra-articularly then intravenously 1×10^{10} PFU/mL (10 mL, IA), 2 days 1×10^{10} PFU/mL (50 mL, IV), 4 days | Daptomycin | Resolution | Not reported | [69] |
| Doub et al., 2022 | Chronic PJI | 1 | MRSA | Personalized phage therapy | Intra-articularly then intravenously 1×10^9 PFU/mL (10 mL, IA), single dose 2×10^8 PFU/mL, 3 doses, IV | Daptomycin Ceftaroline | Resolution | Increased liver enzymes | [70] |

Continued on next page

Table I. Clinical use of bacteriophage therapy (continued)

| Author, year | Infection | n | Microbiology | Phage preparation | Route of administration, dosage, frequency and duration | Combined antibiotics | Reported outcome | Adverse events | Ref. |
|------------------------|---|---|--|----------------------------|--|--|--|----------------------------|------|
| Doub et al., 2022 | Chronic PJI | 1 | <i>Klebsiella pneumoniae</i> ESBL + | Personalized phage therapy | Intra-articularly then intravenously 1×10^{10} PFU/mL (20 mL, IA), 2 days 1×10^{10} PFU/mL (50 mL, IV), 2 days | Ertapenem | Resolution | Not reported | [71] |
| Ferry et al., 2022 | Spondylodiscitis with spinal abscess | 1 | <i>Pseudomonas aeruginosa</i> | Personalized phage therapy | Intraoperative and intravenously 1×10^6 PFU/mL (7 mL) IO, single dose 1×10^6 PFU/mL (30 mL) IV, 21 days | Cefiderocol Colistin | Resolution | No adverse events reported | [72] |
| Schoeffel et al., 2022 | Prosthetic joint infection | 1 | MRSA | Personalized phage therapy | Intra-articularly or intravenously 1.2×10^9 PFU/mL (10 mL), IA, single dose 1.2×10^9 PFU/mL, IV, 3 days | Daptomycin Trimethoprim/ sulfamethoxazole | Resolution | Increased liver enzymes | [73] |
| Eskenazi et al., 2022 | Osteoarticular infection related to left femur fracture | 1 | <i>Klebsiella pneumoniae</i> MSSA | Personalized phage therapy | Intraoperative and catheter drainage 1×10^8 PFU/mL (100 mL) IO 1×10^8 PFU/mL (20 mL) CD, q8h, 5 days | Meropenem Colistin Ceftazidime/ avibactam | Resolution | Not reported | [74] |
| Racenis et al., 2022 | Periprosthetic joint infection and chronic osteomyelitis | 1 | MDR <i>Pseudomonas aeruginosa</i> | Personalized phage therapy | Intraoperative and catheter drainage 1×10^9 PFU/mL (100 mL) IO 1×10^8 PFU/mL (20 mL) CD, q8h, 14 days | Colistin Meropenem Ceftazidime | Resolution | Not reported | [75] |
| Neuts et al., 2021 | Osteoarticular/recurrent chronic infection Prosthetic Hip Infection | 1 | <i>Enterococcus faecalis</i> | Commercial phage cocktail | Oral (dose not reported), twice daily Two 19-day cycles separated by a 2-week break (no surgery) | Amoxicillin Doxycycline | Resolution | Not reported | [76] |
| Ferry et al., 2021 | Osteoarticular Prosthetic Knee Infection | 1 | <i>Pseudomonas aeruginosa</i> | Personalized phage therapy | Intra-articularly 1×10^9 PFU/mL (30 mL) Single dose | Ceftazidime Ciprofloxacin | Resolution (death not related to infection) | Not reported | [77] |

Continued on next page

Table I. Clinical use of bacteriophage therapy (continued)

| Author, year | Infection | <i>n</i> | Microbiology | Phage preparation | Route of administration, dosage, frequency and duration | Combined antibiotics | Reported outcome | Adverse events | Ref. |
|-------------------------------|--|----------|---|----------------------------|--|--|------------------|---|------|
| Doub et al., 2021 | Osteoarticular Prosthetic Knee Infection | 1 | MRSE | Personalized phage therapy | Intra-articularly 2×10^{10} PFU (10 mL) Single dose | Daptomycin | Resolution | Increased liver enzymes | [78] |
| Cano EJ et al., 2021 | Osteoarticular PJI | 1 | <i>Klebsiella pneumoniae</i> | Personalized phage therapy | Intravenously 6.3×10^{10} PFU (50 mL) Once daily, 8 weeks | Minocycline | Resolution | Not reported | [79] |
| Ramirez-Sanchez et al., 2021 | Prosthetic Joint Infection | 1 | MSSA | Personalized phage therapy | Intra-articularly and intravenously 1×10^{10} PFU/mL (50 mL) IA, single dose 1×10^{10} PFU/mL (30 mL) IV, BID, 6 weeks | Cefazoline | Resolution | Serum neutralization | [80] |
| Van Nieuwenhuyse et al., 2021 | Chronic polymicrobial infection of pelvic allografts | 1 | <i>Clostridium hathewayi</i> , <i>Finegoldia magna</i> , <i>Proteus mirabilis</i> , <i>S. aureus</i> | Personalized phage therapy | Intraoperative and catheter drainage 1×10^7 PFU/mL (50 mL) IO, single dose 1×10^7 PFU/mL (40 mL) CD, TID, 7 days 1×10^7 PFU/mL (30 mL) CD, BID, 7 days | Clindamycin Rifampicin Ciprofloxacin | Resolution | Not reported | [81] |
| Ferry et al., 2020 | Prosthetic Knee Infection | 3 | MSSA | Personalized phage therapy | Intra-articularly 1×10^9 PFU/mL, IA, single dose | Daptomycin-based combination, followed by oral therapy and SAT | Resolution | Not reported | [82] |
| Doub et al., 2020 | Chronic periprosthetic infection | 1 | MRSA | Personalized phage therapy | Intra-articularly and intravenously 5.4×10^9 PFU/mL (10 mL) IA, single dose 2.7×10^9 PFU/mL (50 mL) IV, 3 days | Daptomycin | Resolution | Increased liver enzymes (IV phage therapy discontinued) | [83] |

Continued on next page

Table I. Clinical use of bacteriophage therapy (continued)

| Author, year | Infection | n | Microbiology | Phage preparation | Route of administration, dosage, frequency and duration | Combined antibiotics | Reported outcome | Adverse events | Ref. |
|--|--|---|--|--|---|--|---|--|------|
| Onsea et al., 2019 | Chronic osteomyelitis | 4 | <i>S. epidermidis</i> <i>Pseudomonas aeruginosa</i> <i>S. aureus</i> <i>Enterococcus faecalis</i> | Personalized phage therapy (one patient with <i>P. aeruginosa</i> was non-susceptible to phage); Commercial phage cocktail (for <i>E. faecalis</i>) | Intraoperative and catheter drainage 1×10^7 (10–40 mL) IO, single dose 1×10^7 (10–40 mL) CD, TID, 7–10 days | Concomitant systemic antibiotics (including glycopeptides \pm rifampicin), adjusted to pathogen susceptibility | Resolution (one patient had recurrence of infection, although with a different agent) | One patient with local pain and redness (patient with the commercial phage cocktail) | [84] |
| Nir-Paz et al., 2019 | Bone traumatic left tibial infection | 1 | <i>Acinetobacter baumannii</i> , <i>Klebsiella pneumoniae</i> | Personalized phage therapy | Intravenously 5×10^7 PFU/mL (1 mL), TID, 5 days | Colistin Meropenem | Resolution | Not reported | [85] |
| Ferry et al., 2018 | Chronic periprosthetic infection | 1 | <i>S. aureus</i> | Personalized phage therapy | Intra-articularly 1×10^{10} PFU/mL (10 mL), single dose | Daptomycin Amoxicillin Clindamycin | Resolution | Not reported | [86] |
| Ferry et al., 2018 | Periprosthetic infection | 1 | XDR <i>Pseudomonas aeruginosa</i> | Personalized phage therapy | Intra-articularly $\approx 10^8$ PFU/mL (10 mL), every 3 days, 4 administrations | Ceftolozane–tazobactam (local: colistin) | Resolution | Not reported | [87] |
| Fish et al., 2018 | Diabetic foot ulcer with osteomyelitis | 1 | MRSA | Commercial phage cocktail | Local perilesional soft-tissue and intraosseous injection 0.7 mL (PFU not reported), once weekly, 7 weeks | Levofloxacin (without any improvement and suspended) | Resolution | Not reported | [88] |
| Cardiovascular and intravascular infections | | | | | | | | | |
| Eifferman et al., 2025 | Recurrent prosthetic valve endocarditis with infected Bentall (aortic graft infection) | 1 | <i>P. aeruginosa</i> | Personalized phage therapy | Intravenously 1×10^{10} PFU (10 mL), BID, 7 days | Ceftazidime Ciprofloxacin | Resolution | Not reported | [89] |

Continued on next page

Table I. Clinical use of bacteriophage therapy (continued)

| Author, year | Infection | n | Microbiology | Phage preparation | Route of administration, dosage, frequency and duration | Combined antibiotics | Reported outcome | Adverse events | Ref. |
|------------------------|--|---|--|----------------------------|--|--|------------------------------------|----------------|------|
| Hameed et al., 2024 | Post-cardiac surgery mediastinitis | 1 | MDR <i>Klebsiella pneumoniae</i> | Personalized phage therapy | Local administration via mediastinal drains | Systemic antibiotics maintained (not fully detailed) | Resolution | Not reported | [90] |
| Racenis et al., 2023 | LVAD driveline infection | 1 | MDR <i>Pseudomonas aeruginosa</i> | Personalized phage therapy | IV + local; IV concentrations 10^7 – 10^{11} PFU/mL | Colistin Ceftazidime/ avibactam Amikacin | Resolution | Not reported | [91] |
| Rojas et al., 2022 | Intrapericardial LVAD outflow graft infection | 1 | <i>Staphylococcus aureus</i> | Personalized phage therapy | Local application at wound closure | Systemic antibiotics maintained (not fully detailed) | Significant reduction of infection | Not reported | [92] |
| Grambow et al., 2022 | Infected thoracic aortic stent graft | 1 | MSSA | Personalized phage therapy | Local extravascular + endovascular application, repeated instillation over 3 days | Flucloxacillin Cefuroxime | Resolution | Not reported | [93] |
| Puschel et al., 2022 | LVAD driveline infection | 1 | <i>Proteus mirabilis</i> <i>S. aureus</i> | Personalized phage therapy | Local application after debridement | Piperacillin tazobactam | Resolution | Not reported | [94] |
| Rubalskii et al., 2020 | Implant- and transplant-associated cardiothoracic infections | 8 | <i>S. aureus</i> , <i>E. faecium</i> , <i>Pseudomonas aeruginosa</i> , <i>Klebsiella pneumoniae</i> , <i>E. coli</i> | Personalized phage therapy | Variable: local ± oral ± inhaled $\sim 1 \times 10^8$ PFU/mL Duration not standardized | Systemic antibiotics maintained (not fully detailed) | Eradication in 7/8 patients | Not reported | [95] |
| Aslam et al., 2019 | LVAD infection with sternal osteomyelitis and recurrent bacteremia | 1 | MSSA | Personalized phage therapy | Intravenously 3×10^9 PFU BID 28 days | Cefazolin Minocycline | Resolution | Not reported | [96] |

Continued on next page

Table I. Clinical use of bacteriophage therapy (continued)

| Author, year | Infection | n | Microbiology | Phage preparation | Route of administration, dosage, frequency and duration | Combined antibiotics | Reported outcome | Adverse events | Ref. |
|-------------------------------------|---|---|--|----------------------------|---|---|---|--|-------|
| Gilbey et al., 2019 | Prosthetic valve endocarditis | 1 | MSSA | Personalized phage therapy | Intravenously 1×10^9 PFU BID 14 days | Flucloxacillin + ciprofloxacin + rifampicin | Resolution | No adverse event attributable to phage therapy | [97] |
| Chan et al., 2018 | Aortic graft infection with aorto-cutaneous fistula | 1 | <i>Pseudomonas aeruginosa</i> | Personalized phage therapy | Single local application directly to graft | Ceftazidime | Resolution | Not reported | [98] |
| Respiratory tract infections | | | | | | | | | |
| Yang et al., 2025 | Lung infection | 1 | Carbapenem-resistant <i>Pseudomonas aeruginosa</i> | Personalized phage therapy | Nebulized 9×10^9 PFU/mL 14 days | Colistin | Clinical improvement; persistent <i>Pseudomonas aeruginosa</i> colonization (no eradication). Phage resistance detected | Not reported | [99] |
| Tenney et al., 2024 | VAP (burned patient) | 1 | XDR <i>Pseudomonas aeruginosa</i> | Personalized phage therapy | Nebulized and intravenous 2×10^9 PFU/mL, 3 inhaled doses, 5 mL 2×10^8 PFU/mL, daily IV, 50 mL, 7 days | Imipenem–relebactam Colistin (inhaled) | Resolution (pneumonia recurrence after 1 month; repeated phage course with favorable outcome) | No adverse event attributable to phage therapy | [100] |

Continued on next page

Table I. Clinical use of bacteriophage therapy (continued)

| Author, year | Infection | n | Microbiology | Phage preparation | Route of administration, dosage, frequency and duration | Combined antibiotics | Reported outcome | Adverse events | Ref. |
|----------------------|--|---|-------------------------------------|----------------------------|--|---|--|--|-------|
| Kohler et al., 2023 | Lung infection | 1 | MDR <i>Pseudomonas aeruginosa</i> | Personalized phage therapy | Nebulized daily dose of 5×10^9 PFU, 7 days | Concomitant suppressive antibiotics (not specified) | Clinical improvement without eradication; recurrence requiring repeated phage course | Transient hypoxia, single febrile episode | [101] |
| Li et al., 2023 | Chronic lung infection (interstitial lung disease) | 1 | <i>Pseudomonas aeruginosa</i> | Personalized phage therapy | Nebulized 1×10^8 PFU/mL (10 mL) BID, 3 days | Multiple systemic antibiotics prior to phage therapy, discontinued during repeated nebulized phage courses | Transient elimination of <i>Pseudomonas aeruginosa</i> in sputum; significant improvement of infection; recurrence, not eradicated | Not reported | [102] |
| Haidar et al., 2023 | Lung infection (lung transplant) | 1 | MDR <i>Burkholderia multivorans</i> | Not specified | Nebulized 3.33×10^9 PFU per dose 3 mL per dose 7 days | Concomitant suppressive antibiotics (not specified) | Clinical failure and death | No adverse event attributable to phage therapy | [103] |
| Levêque et al., 2023 | Lung infection (lung transplant) | 1 | XDR <i>Pseudomonas aeruginosa</i> | Personalized phage therapy | Multiple nebulized doses (not specified) | Continuous inhaled colistin and multiple systemic antibiotics (including cefiderocol and aminoglycosides, adjusted over time) | Microbiological response without clinical success; death | Not reported | [104] |

Continued on next page

Table I. Clinical use of bacteriophage therapy (continued)

| Author, year | Infection | n | Microbiology | Phage preparation | Route of administration, dosage, frequency and duration | Combined antibiotics | Reported outcome | Adverse events | Ref. |
|-----------------------|---|---|--|----------------------------|--|---|--|----------------|-------|
| Li et al., 2023 | Chronic pulmonary infection | 1 | MDR <i>Klebsiella pneumoniae</i> | Personalized phage therapy | Nebulized > 1 × 10 ⁹ PFU/mL 5 mL, 2 single-day courses, each with 3 nebulized doses | Concomitant systemic antibiotics (regimens not specified) | Clinical improvement despite incomplete eradication. Phage resistance detected | Not reported | [105] |
| Chen et al., 2022 | Chronic pleuropulmonary infection after lobectomy | 1 | MDR <i>Pseudomonas aeruginosa</i> | Personalized phage therapy | Nebulized and intrapleural Dosage not consistently reported Nebulized BID and intrapleural QD, 11 days and then intensified nebulized regimen (duration not specified) | Amikacin Ceftazidime–avibactam Fosfomycin | Persistent colonization; increased susceptibility to antibiotics after phage therapy | Not reported | [106] |
| Wu et al., 2021 | Secondary bacterial pneumonia | 4 | Carbapenem-resistant <i>Acinetobacter baumannii</i> | Personalized phage therapy | Nebulized 2 × 10 ⁹ PFU per administration Repeated doses (exact duration varied per patient) | Concomitant suppressive antibiotics (not specified) | Reduced bacterial load with clinical improvement | Not reported | [107] |
| Maddocks et al., 2019 | VAP | 1 | MDR <i>Pseudomonas aeruginosa</i> | Personalized phage therapy | Nebulized and intravenously 4 mL undiluted, BID, Neb 100 mL, BID, IV | Ciprofloxacin Gentamicin | Resolution | Not reported | [108] |
| Aslam et al., 2019 | Lung transplant | 3 | <i>Pseudomonas aeruginosa</i> (n=2), <i>Burkholderia dolosa</i> (n=1) | Personalized phage therapy | Nebulized and intravenously (exact PFU and duration not specified) | Concomitant suppressive antibiotics (not specified) | Clinical improvement in 2/3 patients; relapse and death in 1 patient | Not reported | [109] |
| Law et al., 2019 | Lung infection (waiting lung transplant) | 1 | MDR <i>Pseudomonas aeruginosa</i> | Personalized phage therapy | Intravenously 1 × 10 ⁹ PFU/mL (IV dosing, repeated) | Colistin Azithromycin Piperacillin–tazobactam Carbapenem | Resolution | Not reported | [110] |

Continued on next page

Table I. Clinical use of bacteriophage therapy (continued)

| Author, year | Infection | <i>n</i> | Microbiology | Phage preparation | Route of administration, dosage, frequency and duration | Combined antibiotics | Reported outcome | Adverse events | Ref. |
|--|-------------------------------|----------|-------------------------------------|--|---|-------------------------|---|----------------|-------|
| Genito-Urinary Tract Infections | | | | | | | | | |
| Cook et al., 2025 | Recurrent UTI | 1 | MDR <i>E. coli</i> | Personalized phage therapy | Oral, intravesical instillation and then topical formulation 4.3×10^{11} PFU in 100 mL, single dose, oral 4.3×10^{11} PFU in 100 mL, single dose 2.6×10^9 PFU (topical), 3 days | Not specified | Clinical improvement with reduction in UTI episodes | Not reported | [111] |
| Cesta et al., 2025 | Chronic bacterial prostatitis | 1 | <i>E. coli</i> | Personalized phage therapy | Rectal and oral administration for several weeks (exact PFU and duration not specified) | No combined antibiotics | Resolution | Not reported | [112] |
| Johri et al., 2023 | Chronic bacterial prostatitis | 1 | <i>E. coli</i> | Personalized phage therapy | Rectal and oral administration for several weeks (exact PFU and duration not specified) | No combined antibiotics | Resolution | Not reported | [113] |
| Le et al., 2023 | Recurrent UTI (transplant) | 1 | <i>Klebsiella pneumoniae</i> ESBL + | Personalized phage therapy | Intravenously, 4 weeks (exact PFU not specified) | No combined antibiotics | Resolution, no recurrence for 6 months | Not reported | [114] |
| Johri et al., 2021 | Chronic prostatitis | 1 | <i>E. coli</i> | Personalized phage therapy | Oral and rectal Several weeks (exact PFU and duration not specified) | No combined antibiotics | Resolution | Not reported | [115] |
| Rostkowska et al., 2021 | Recurrent UTI | 1 | <i>Klebsiella pneumoniae</i> ESBL + | Phage cocktail with confirmed lytic activity | Intrarectal, repeated courses (exact PFU and duration not specified) | Meropenem Colistin | Resolution only after nephrectomy | Not reported | [116] |
| Terwilliger et al., 2021 | Recurrent UTI and prostatitis | 1 | <i>E. coli</i> ESBL + | Personalized phage therapy | Intravenous, 2 weeks (exact PFU not specified) | Ertapenem | Resolution | Not reported | [117] |
| Bao et al., 2020 | UTI | 1 | <i>Klebsiella pneumoniae</i> | Personalized phage therapy | Intravesical, repeated instillations (exact PFU and duration not specified) | Not clearly specified | Reduction in bacterial burden and symptom improvement | Not reported | [118] |

Continued on next page

Table I. Clinical use of bacteriophage therapy (continued)

| Author, year | Infection | <i>n</i> | Microbiology | Phage preparation | Route of administration, dosage, frequency and duration | Combined antibiotics | Reported outcome | Adverse events | Ref. |
|-------------------------|----------------------------------|----------|--|--|---|-------------------------|------------------|----------------|-------|
| Kuipers et al., 2019 | UTI (renal transplant recipient) | 1 | <i>Klebsiella pneumoniae</i> ESBL + | Phage cocktail with confirmed lytic activity | Oral and intravesical, 12 weeks total (exact PFU and duration not specified) | Meropenem | Resolution | Not reported | [119] |
| Khawaldeh et al., 2011 | Recurrent UTI | 1 | <i>Pseudomonas aeruginosa</i> | Personalized phage therapy | Intravesical, 10 days (exact PFU and duration not specified) | Meropenem Colistin | Resolution | Not reported | [120] |
| Letkiewicz et al., 2009 | Chronic bacterial prostatitis | 3 | <i>Enterococcus faecalis</i> | Personalized phage therapy | Rectal; 2.8×10^8 / 7.5×10^7 / 4.5×10^7 PFU/mL (10 mL) BID 28–33 days | No combined antibiotics | Resolution | Not reported | [121] |

Notes: (1) Phage preparation terminology: **Personalized phage therapy** indicates bacteriophages specifically selected, adapted, or produced for an individual patient following susceptibility testing. **Commercial phage cocktail** denotes standardized phage products administered without documented patient-specific selection. **Phage cocktail with confirmed lytic activity** describes phage preparations obtained from established phage banks or pre-existing cocktails, for which in vitro lytic activity against the clinical isolate was confirmed before administration.

Abbreviations. BID, twice daily; CD, catheter drainage; ESBL, extended-spectrum beta-lactamase; IA, intra-articular; IO, intraoperative; IV, intravenous; LVAD, left ventricular assist device; MDR, multidrug-resistant; MRSA, methicillin-resistant *Staphylococcus aureus*; MRSE, methicillin-resistant *Staphylococcus epidermidis*; MSSA, methicillin-susceptible *Staphylococcus aureus*; Neb, nebulized; PFU, plaque-forming units; PJI, prosthetic joint infection; QD, once daily; SAT, suppressive antibiotic therapy; TID, three times daily; UTI, urinary tract infection; VAP, ventilator-associated pneumonia; XDR, extensively drug-resistant.

References

1. Twort FW. An investigation on the nature of ultra-microscopic viruses. *Bacteriophage*. 2011;1(3):127–129. <https://doi.org/10.4161/bact.1.3.16737>
2. D'Herelle F. On an invisible microbe antagonistic toward dysenteric bacilli: brief note by Mr. F. D'Herelle, presented by Mr. Roux. *Res Microbiol*. 2007;158(7):553–554. <https://doi.org/10.1016/j.resmic.2007.07.005>
3. World Health Organization. *WHO Global Antimicrobial Resistance and Use Surveillance System (GLASS): global antibiotic resistance surveillance report*. Geneva: World Health Organization; 2025.
4. Kortright KE, Chan BK, Koff JL, Turner PE. Phage therapy: a renewed approach to combat antibiotic-resistant bacteria. *Cell Host Microbe*. 2019;25(2):219–232. <https://doi.org/10.1016/j.chom.2019.01.014>
5. Grincho N, Mateus F, Silva P, Sousa H. Resistências aos antimicrobianos: flagelo socioeconómico do século XXI. *Revista da UI IPSantarém*. 2019;7(2):112–114. <https://doi.org/10.25746/ruiips.v7.i2.19303>
6. Clokie MRJ, Millard AD, Letarov AV, Heaphy S. Phages in nature. *Bacteriophage*. 2011;1(1):31–45. <https://doi.org/10.4161/bact.1.1.14942>
7. Fernández L, Gutiérrez D, García P, Rodríguez A. The perfect bacteriophage for therapeutic applications— a quick guide. *Antibiotics*. 2019;8(3):126. <https://doi.org/10.3390/antibiotics8030126>
8. Comeau AM, Hatfull GF, Krisch HM, Lindell D, Mann NH, Prangishvili D. Exploring the prokaryotic virosphere. *Res Microbiol*. 2008;159(5):306–313. <https://doi.org/10.1016/j.resmic.2008.05.001>
9. Turner D, Shkorporov AN, Lood C, Millard AD, Dutilh BE, Alfenas-Zerbini P, et al. Abolishment of morphology-based taxa and change to binomial species names: 2022 taxonomy update of the ICTV bacterial viruses subcommittee. *Arch Virol*. 2023;168(2):74. <https://doi.org/10.1007/s00705-022-05694-2>
10. Velesler D, Cambillau C. A common evolutionary origin for tailed-bacteriophage functional modules and bacterial machineries. *Microbiol Mol Biol Rev*. 2011;75(3):423–433. <https://doi.org/10.1128/MMBR.00014-11>
11. Salmond GPC, Fineran PC. A century of the phage: past, present and future. *Nat Rev Microbiol*. 2015;13(12):777–786. <https://doi.org/10.1038/nrmiicro3564>
12. Ofir G, Sorek R. Contemporary phage biology: from classic models to new insights. *Cell*. 2018;172(6):1260–1270. <https://doi.org/10.1016/j.cell.2017.10.045>
13. Gummalla VS, Zhang Y, Liao Y-T, Wu VCH. The role of temperate phages in bacterial pathogenicity. *Microorganisms*. 2023;11(3):541. <https://doi.org/10.3390/microorganisms11030541>
14. Schooley RT, Biswas B, Gill JJ, Hernandez-Morales A, Lancaster J, Lessor L, 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(10):e00954-17. <https://doi.org/10.1128/AAC.00954-17>
15. Hyman P, Abedon ST. Bacteriophage host range and bacterial resistance. In: *Advances in Applied Microbiology*. Vol 70. 2010. p. 217–248. [https://doi.org/10.1016/S0065-2164\(10\)70007-1](https://doi.org/10.1016/S0065-2164(10)70007-1)
16. Olawade DB, Fapohunda O, Egbon E, Ebiesuwa OA, Usman SO, Faronbi AO, et al. Phage therapy: a targeted approach to overcoming antibiotic resistance. *Microb Pathog*. 2024;197:107088. <https://doi.org/10.1016/j.micpath.2024.107088>
17. Labrie SJ, Samson JE, Moineau S. Bacteriophage resistance mechanisms. *Nat Rev Microbiol*. 2010;8(5):317–327. <https://doi.org/10.1038/nrmicro2315>
18. Bernheim A, Sorek R. The pan-immune system of bacteria: antiviral defence as a community resource. *Nat Rev Microbiol*. 2020;18(2):113–119. <https://doi.org/10.1038/s41579-019-0278-2>
19. Rohde C, Resch G, Pirnay J-P, Blasdel B, Debarbieux L, Gelman D, et al. Expert opinion on three phage therapy related topics: bacterial phage resistance, phage training and prophages in bacterial production strains. *Viruses*. 2018;10(4):178. <https://doi.org/10.3390/v10040178>
20. Hall AR, De Vos D, Friman V-P, Pirnay J-P, Buckling A. Effects of sequential and simultaneous applications of bacteriophages on populations of *Pseudomonas aeruginosa* in vitro and in wax moth larvae. *Appl Environ Microbiol*. 2012;78(16):5646–5652. <https://doi.org/10.1128/AEM.00757-12>
21. Mangalea MR, Duerkop BA. Fitness trade-offs resulting from bacteriophage resistance potentiate synergistic antibacterial strategies. *Infect Immun*. 2020;88(4):e00926-19. <https://doi.org/10.1128/IAI.00926-19>
22. Egado JE, Costa AR, Aparicio-Maldonado C, Haas P-J, Brouns SJJ. Mechanisms and clinical importance of bacteriophage resistance. *FEMS Microbiol Rev*. 2022;46(1):fuab048. <https://doi.org/10.1093/femsre/fuab048>
23. Sauer K, Stoodley P, Goeres DM, Hall-Stoodley L, Burmølle M, Stewart PS, et al. The biofilm life cycle: expanding the conceptual model of biofilm formation. *Nat Rev Microbiol*. 2022;20(10):608–620. <https://doi.org/10.1038/s41579-022-00767-0>

24. Harper DR, Parracho HMRT, Walker J, Sharp R, Hughes G, Werthén M, et al. Bacteriophages and biofilms. *Antibiotics*. 2014;3(3):270–284. <https://doi.org/10.3390/antibiotics3030270>
25. Pires DP, Melo LDR, Vilas Boas D, Sillankorva S, Azeredo J. Phage therapy as an alternative or complementary strategy to prevent and control biofilm-related infections. *Curr Opin Microbiol*. 2017;39:48–56. <https://doi.org/10.1016/j.mib.2017.09.004>
26. Meneses L, Brandão AC, Coenye T, Braga AC, Pires DP, Azeredo J. A systematic review of the use of bacteriophages for in vitro biofilm control. *Eur J Clin Microbiol Infect Dis*. 2023;42(8):919–928. <https://doi.org/10.1007/s10096-023-04638-1>
27. Turner PE, Azeredo J, Buurman ET, Green S, Haaber JK, Haggstrom D, et al. Addressing the research and development gaps in modern phage therapy. *PHAGE*. 2024;5(1):30–39. <https://doi.org/10.1089/phage.2023.0045>
28. Comeau AM, Tétart F, Trojet SN, Prère M-F, Krisch HM. Phage-antibiotic synergy (PAS): β -lactam and quinolone antibiotics stimulate virulent phage growth. *PLoS One*. 2007;2(8):e799. <https://doi.org/10.1371/journal.pone.0000799>
29. Gordillo Altamirano FL, Barr JJ. Phage therapy in the postantibiotic era. *Clin Microbiol Rev*. 2019;32(2):e00066–18. <https://doi.org/10.1128/CMR.00066-18>
30. Morrisette T, Kebriaei R, Lev K, Morales S, Rybak MJ. Bacteriophage therapeutics: a primer for clinicians on phage-antibiotic combinations. *Pharmacotherapy*. 2020;40(2):153–168. <https://doi.org/10.1002/phar.2358>
31. Supina BSI, Dennis JJ. The current landscape of phage-antibiotic synergistic (PAS) interactions. *Antibiotics*. 2025;14(6):545. <https://doi.org/10.3390/antibiotics14060545>
32. Diallo K, Dublanche A. Benefits of combined phage-antibiotic therapy for the control of antibiotic-resistant bacteria: a literature review. *Antibiotics*. 2022;11(7):839. <https://doi.org/10.3390/antibiotics11070839>
33. Summers WC. Bacteriophage therapy. *Annu Rev Microbiol*. 2001;55:437–451. <https://doi.org/10.1146/annurev.micro.55.1.437>
34. Lin DM, Koskella B, Lin HC. Phage therapy: an alternative to antibiotics in the age of multi-drug resistance. *World J Gastrointest Pharmacol Ther*. 2017;8(3):162–173. <https://doi.org/10.4292/wjgpt.v8.i3.162>
35. Łusiak-Szelachowska M, Weber-Dąbrowska B, Górski A. Bacteriophages and lysins in biofilm control. *Viol Sin*. 2020;35(2):125–133. <https://doi.org/10.1007/s12250-019-00192-3>
36. Kever L, Hardy A, Luthe T, Hünnefeld M, Gätgens C, Milke L, et al. Aminoglycoside antibiotics inhibit phage infection by blocking an early step of the infection cycle. *mBio*. 2022;13(2):e00783–22. <https://doi.org/10.1128/mbio.00783-22>
37. Abedon ST. Phage-antibiotic combination treatments: antagonistic impacts of antibiotics on the pharmacodynamics of phage therapy? *Antibiotics*. 2019;8(4):182. <https://doi.org/10.3390/antibiotics8040182>
38. Jiang Z, Wei J, Liang Y, Peng N, Li Y. Aminoglycoside antibiotics inhibit mycobacteriophage infection. *Antibiotics*. 2020;9(10):714. <https://doi.org/10.3390/antibiotics9100714>
39. Dimitriu T, Kurilovich E, Łapińska U, Severinov K, Pagliara S, Szczelkun MD, et al. Bacteriostatic antibiotics promote CRISPR-Cas adaptive immunity by enabling increased spacer acquisition. *Cell Host Microbe*. 2022;30(1):31–40.e5. <https://doi.org/10.1016/j.chom.2021.11.014>
40. Hille F, Richter H, Wong SP, Bratovič M, Ressel S, Charpentier E. The biology of CRISPR-Cas: backward and forward. *Cell*. 2018;172(6):1239–1259. <https://doi.org/10.1016/j.cell.2017.11.032>
41. Liu H, Li H, Liang Y, Du X, Yang C, Yang L, et al. Phage-delivered sensitisation with subsequent antibiotic treatment reveals sustained effect against antimicrobial resistant bacteria. *Theranostics*. 2020;10(14):6310–6321. <https://doi.org/10.7150/thno.42573>
42. Huh K, Chung DR, Ha YE, Ko J-H, Kim S-H, Kim M-J, et al. Impact of difficult-to-treat resistance in Gram-negative bacteremia on mortality: retrospective analysis of nationwide surveillance data. *Clin Infect Dis*. 2020;71(9):e487–e496. <https://doi.org/10.1093/cid/ciaa084>
43. Sati H, Carrara E, Savoldi A, Hansen P, Garlasco J, Campagnaro E, et al.; WHO Bacterial Priority Pathogens List Advisory Group. The WHO Bacterial Priority Pathogens List 2024: a prioritisation study to guide research, development, and public health strategies against antimicrobial resistance. *Lancet Infect Dis*. 2025;25(9):1033–1043. [https://doi.org/10.1016/S1473-3099\(25\)00118-5](https://doi.org/10.1016/S1473-3099(25)00118-5)
44. Cisek AA, Dąbrowska I, Gregorczyk KP, Wyżewski Z. Phage therapy in bacterial infections treatment: one hundred years after the discovery of bacteriophages. *Curr Microbiol*. 2017;74(2):277–283. <https://doi.org/10.1007/s00284-016-1166-x>
45. Pires DP, Vilas Boas D, Sillankorva S, Azeredo J. Phage therapy: a step forward in the treatment of *Pseudomonas aeruginosa* infections. *J Virol*. 2015;89(15):7449–7456. <https://doi.org/10.1128/jvi.00385-15>

46. Moller AG, Winston K, Ji S, Wang J, Hargita Davis MN, Solís-Lemus CR, et al. Genes influencing phage host range in *Staphylococcus aureus* on a species-wide scale. *mSphere*. 2021;6(1):e01263-20. <https://doi.org/10.1128/mSphere.01263-20>
47. Haudiquet M, Le Bris J, Nucci A, Bonnin RA, Domingo-Calap P, Rocha EPC, et al. Capsules and their traits shape phage susceptibility and plasmid conjugation efficiency. *Nat Commun*. 2024;15:2032. <https://doi.org/10.1038/s41467-024-46147-5>
48. Bai J, Raustad N, Denoncourt J, van Opijnen T, Geisinger E. Genome-wide phage susceptibility analysis in *Acinetobacter baumannii* reveals capsule modulation strategies that determine phage infectivity. *PLoS Pathog*. 2023;19(3):e1010928. <https://doi.org/10.1371/journal.ppat.1010928>
49. Resch G, Brives C, Debarbieux L, Hodges FE, Kirchhelle C, Laurent F, et al. Between centralization and fragmentation: the past, present, and future of phage collections. *PHAGE*. 2024;5(1):22-29. <https://doi.org/10.1089/phage.2023.0043>
50. Nagel T, Musila L, Muthoni M, Nikolich M, Nakavuma JL, Clokie MRJ. Phage banks as potential tools to rapidly and cost-effectively manage antimicrobial resistance in the developing world. *Curr Opin Virol*. 2022;53:101208. <https://doi.org/10.1016/j.coviro.2022.101208>
51. Żaczek M, Górski A, Weber-Dąbrowska B, Letkiewicz S, Fortuna W, Rogóż P, et al. A thorough synthesis of phage therapy unit activity in Poland—its history, milestones and international recognition. *Viruses*. 2022;14(6):1170. <https://doi.org/10.3390/v14061170>
52. Malik DJ, Sokolov IJ, Vinner GK, Mancuso F, Cinquerrui S, Vladislavljevic GT, et al. Formulation, stabilisation and encapsulation of bacteriophage for phage therapy. *Adv Colloid Interface Sci*. 2017;249:100-133. <https://doi.org/10.1016/j.cis.2017.05.014>
53. Bonilla N, Barr JJ. Phage on tap: a quick and efficient protocol for the preparation of bacteriophage laboratory stocks. *Methods Mol Biol*. 2018;1838:37-46. https://doi.org/10.1007/978-1-4939-8682-8_4
54. Sulakvelidze A, Alavidze Z, Morris JG Jr. Bacteriophage therapy. *Antimicrob Agents Chemother*. 2001;45(3):649-659. <https://doi.org/10.1128/AAC.45.3.649-659.2001>
55. Hanlon GW. Bacteriophages: an appraisal of their role in the treatment of bacterial infections. *Int J Antimicrob Agents*. 2007;30(2):118-128. <https://doi.org/10.1016/j.ijantimicag.2007.04.006>
56. Chegini Z, Khoshbayan A, Taati Moghadam M, Farahani I, Jazireian P, Shariati A. Bacteriophage therapy against *Pseudomonas aeruginosa* biofilms: a review. *Ann Clin Microbiol Antimicrob*. 2020;19:45. <https://doi.org/10.1186/s12941-020-00389-5>
57. Pires DP, Melo LDR, Vilas Boas D, Sillankorva S, Azeredo J. Phage therapy as an alternative or complementary strategy to prevent and control biofilm-related infections. *Curr Opin Microbiol*. 2017;39:48-56. <https://doi.org/10.1016/j.mib.2017.09.004>
58. Waters EM, Neill DR, Kaman B, Sahota JS, Clokie MRJ, Winstanley C, et al. Phage therapy is highly effective against chronic lung infections with *Pseudomonas aeruginosa*. *Thorax*. 2017;72(7):666-667. <https://doi.org/10.1136/thoraxjnl-2016-209265>
59. Chhibber S, Kaur T, Kaur S. Co-therapy using lytic bacteriophage and linezolid: effective treatment in eliminating methicillin resistant *Staphylococcus aureus* (MRSA) from diabetic foot infections. *PLoS One*. 2013;8(2):e56022. <https://doi.org/10.1371/journal.pone.0056022>
60. Kifelew LG, Warner MS, Morales S, Vaughan L, Woodman R, Fitridge R, et al. Efficacy of phage cocktail AB-SA01 therapy in diabetic mouse wound infections caused by multidrug-resistant *Staphylococcus aureus*. *BMC Microbiol*. 2020;20:204. <https://doi.org/10.1186/s12866-020-01891-8>
61. Fish R, Kutter E, Wheat G, Blasdel B, Kutateladze M, Kuhl S. Bacteriophage treatment of intransigent diabetic toe ulcers: a case series. *J Wound Care*. 2016;25(7 Suppl):S27-S33. <https://doi.org/10.12968/jowc.2016.25.7.S27>
62. Law N, Logan C, Yung G, Furr CLL, Lehman SM, Morales S, et al. Successful adjunctive use of bacteriophage therapy for treatment of multidrug-resistant *Pseudomonas aeruginosa* infection in a cystic fibrosis patient. *Infection*. 2019;47(4):665-668. <https://doi.org/10.1007/s15010-019-01319-0>
63. McCallin S, Sacher JC, Zheng J, Chan BK. Current state of compassionate phage therapy. *Viruses*. 2019;11(4):343. <https://doi.org/10.3390/v11040343>
64. Pires DP, Costa AR, Pinto G, Meneses L, Azeredo J. Current challenges and future opportunities of phage therapy. *FEMS Microbiol Rev*. 2020;44(6):684-700. <https://doi.org/10.1093/femsre/fuaa017>
65. Pires DP, Meneses L, Brandão AC, Azeredo J. An overview of the current state of phage therapy for the treatment of biofilm-related infections. *Curr Opin Virol*. 2022;53:101209. <https://doi.org/10.1016/j.coviro.2022.101209>
66. Eschweiler J, Fischer C, Migliorini F, Greven J, Mendel T, Kobbe P, et al. Is a bacteriophage approach for musculoskeletal infection management an alternative to conventional therapy? *Life*. 2025;15(10):1534. <https://doi.org/10.3390/life15101534>

67. Cammuso MT, Cook BWM, Cameron DW, Ryan S, Tamayo M, Peters MJ, et al. First use of phage therapy in Canada for the treatment of a life-threatening, multidrug-resistant *Staphylococcus epidermidis* periprosthetic joint infection. *Viruses*. 2025;17(8):1118. <https://doi.org/10.3390/v17081118>
68. Wahl P, Schläppi M, Loganathan A, Uçkay I, Hodel S, Fritz B, et al. Bacteriophage therapy created the necessary conditions for successful antibiotic suppression in a periprosthetic hip joint infection: a case report. *Front Med (Lausanne)*. 2025;12:1564369. <https://doi.org/10.3389/fmed.2025.1564369>
69. Doub JB, Chan B, Johnson AJ. Salphage: salvage bacteriophage therapy for a chronic *Enterococcus faecalis* prosthetic joint infection. *IDCases*. 2023;33:e01854. <https://doi.org/10.1016/j.idcr.2023.e01854>
70. Doub JB, Ng VY, Lee M, Chi A, Lee A, Würstle S, et al. Salphage: salvage bacteriophage therapy for recalcitrant MRSA prosthetic joint infection. *Antibiotics*. 2022;11(5):616. <https://doi.org/10.3390/antibiotics11050616>
71. Doub JB, Shishido A, Srikumaran U, Haskoor J, Tran-Nguyen P, Lee M, et al. Salphage: salvage bacteriophage therapy for a recalcitrant *Klebsiella pneumoniae* prosthetic shoulder infection—a case report. *Acta Orthop*. 2022;93:756–759. <https://doi.org/10.2340/17453674.2022.4579>
72. Ferry T, Kolenda C, Laurent F, Leboucher G, Merabishvili M, Djebara S, et al. Personalized bacteriophage therapy to treat pandrug-resistant spinal *Pseudomonas aeruginosa* infection. *Nat Commun*. 2022;13:4239. <https://doi.org/10.1038/s41467-022-31837-9>
73. Schoeffel J, Wang EW, Gill D, Frackler J, Horne B, Manson T, et al. Successful use of salvage bacteriophage therapy for a recalcitrant MRSA knee and hip prosthetic joint infection. *Pharmaceuticals*. 2022;15(2):177. <https://doi.org/10.3390/ph15020177>
74. Eskenazi A, Lood C, Wubbolts J, Hites M, Balarjshvili N, Leshkasheli L, et al. Combination of pre-adapted bacteriophage therapy and antibiotics for treatment of fracture-related infection due to pandrug-resistant *Klebsiella pneumoniae*. *Nat Commun*. 2022;13:302. <https://doi.org/10.1038/s41467-021-27656-z>
75. Racenis K, Rezevska D, Madelane M, Lavrinovics E, Djebara S, Petersons A, et al. Use of phage cocktail BFC 1.10 in combination with ceftazidime-avibactam in the treatment of multidrug-resistant *Pseudomonas aeruginosa* femur osteomyelitis—a case report. *Front Med (Lausanne)*. 2022;9:851310. <https://doi.org/10.3389/fmed.2022.851310>
76. Neuts AS, Berkhout HJ, Hartog A, Goosen JHM. Bacteriophage therapy cures a recurrent *Enterococcus faecalis* infected total hip arthroplasty? A case report. *Acta Orthop*. 2021;92(6):678–680. <https://doi.org/10.1080/17453674.2021.1968714>
77. Ferry T, Kolenda C, Batailler C, Gaillard R, Gustave CA, Lustig S, et al. Case report: arthroscopic "debridement antibiotics and implant retention" with local injection of personalized phage therapy to salvage a relapsing *Pseudomonas aeruginosa* prosthetic knee infection. *Front Med (Lausanne)*. 2021;8:569159. <https://doi.org/10.3389/fmed.2021.569159>
78. Doub JB, Ng VY, Wilson E, Corsini L, Chan BK. Successful treatment of a recalcitrant *Staphylococcus epidermidis* prosthetic knee infection with intraoperative bacteriophage therapy. *Pharmaceuticals*. 2021;14(3):231. <https://doi.org/10.3390/ph14030231>
79. Cano EJ, Caffisch KM, Bollyky PL, Van Belleghem JD, Patel R, Fackler J, et al. Phage therapy for limb-threatening prosthetic knee *Klebsiella pneumoniae* infection: case report and in vitro characterization of anti-biofilm activity. *Clin Infect Dis*. 2021;73(1):e144–e151. <https://doi.org/10.1093/cid/ciaa705>
80. Ramirez-Sanchez C, Gonzales F, Buckley M, Biswas B, Henry M, Deschenes MV, et al. Successful treatment of *Staphylococcus aureus* prosthetic joint infection with bacteriophage therapy. *Viruses*. 2021;13(6):1182. <https://doi.org/10.3390/v13061182>
81. Van Nieuwenhuysse B, Galant C, Brichard B, Docquier PL, Djebara S, Pirnay JP, et al. A case of in situ phage therapy against *Staphylococcus aureus* in a bone allograft polymicrobial biofilm infection: outcomes and phage-antibiotic interactions. *Viruses*. 2021;13(10):1898. <https://doi.org/10.3390/v13101898>
82. Ferry T, Kolenda C, Batailler C, Gustave CA, Lustig S, Malatray M, et al. Phage therapy as adjuvant to conservative surgery and antibiotics to salvage patients with relapsing *S. aureus* prosthetic knee infection. *Front Med (Lausanne)*. 2020;7:570572. <https://doi.org/10.3389/fmed.2020.570572>
83. Doub JB, Ng VY, Johnson AJ, Slomka M, Fackler J, Horne B, et al. Salvage bacteriophage therapy for a chronic MRSA prosthetic joint infection. *Antibiotics*. 2020;9(5):241. <https://doi.org/10.3390/antibiotics9050241>
84. Onsea J, Soentjens P, Djebara S, Merabishvili M, Depypere M, Spriet I, et al. Bacteriophage application for difficult-to-treat musculoskeletal infections: development of a standardized multidisciplinary treatment protocol. *Viruses*. 2019;11(10):891. <https://doi.org/10.3390/v11100891>
85. Nir-Paz R, Gelman D, Khouri A, Sisson BM, Fackler J, Alkalay-Oren S, et al. Successful treatment of antibiotic-resistant, polymicrobial bone infection with bacteriophages and antibiotics combination. *Clin Infect*

- Dis.* 2019;69(11):2015–2018. <https://doi.org/10.1093/cid/ciz222>
86. Ferry T, Leboucher G, Fevre C, Herry Y, Conrad A, Josse J, et al. Salvage debridement, antibiotics and implant retention (“DAIR”) with local injection of a selected cocktail of bacteriophages: is it an option for an elderly patient with relapsing *Staphylococcus aureus* prosthetic-joint infection? *Open Forum Infect Dis.* 2018;5(11):ofy269. <https://doi.org/10.1093/ofid/ofy269>
 87. Ferry T, Boucher F, Fevre C, Perpoint T, Chateau J, Petitjean C, et al. Innovations for the treatment of a complex bone and joint infection due to XDR *Pseudomonas aeruginosa* including local application of a selected cocktail of bacteriophages. *J Antimicrob Chemother.* 2018;73(10):2901–2903. <https://doi.org/10.1093/jac/dky263>
 88. Fish R, Kutter E, Bryan D, Wheat G, Kuhl S. Resolving digital staphylococcal osteomyelitis using bacteriophage—a case report. *Antibiotics.* 2018;7(4):87. <https://doi.org/10.3390/antibiotics7040087>
 89. Eiferman V, Vion PA, Bleibtreu A. Phage therapy as a rescue treatment for recurrent *Pseudomonas aeruginosa* Bentall infection. *Viruses.* 2025;17(1):123. <https://doi.org/10.3390/v17010123>
 90. Hameed I, Ahmed A, Gandhi S, Nassiri N, Steinbacher DM, Vallabhajosyula P. Successful investigational phage therapy for pan-resistant bacterial mediastinitis following type II hybrid aortic arch replacement. *JACC Case Rep.* 2024;29:102816. <https://doi.org/10.1016/j.jaccas.2024.102816>
 91. Racenis K, Lacis J, Rezevska D, Mukane L, Vilde A, Putnins I, et al. Successful bacteriophage-antibiotic combination therapy against multidrug-resistant *Pseudomonas aeruginosa* left ventricular assist device driveline infection. *Viruses.* 2023;15(5):1210. <https://doi.org/10.3390/v15051210>
 92. Rojas SV, Junghans S, Fox H, Lazowski K, Schramm R, Morshuis M, et al. Bacteriophage-enriched galenic for intrapericardial ventricular assist device infection. *Antibiotics.* 2022;11(5):602. <https://doi.org/10.3390/antibiotics11050602>
 93. Grambow E, Junghans S, Kröger JC, Reisinger EC, Krause BJ, Groß J. Treatment of an infected TEVAR with extra- and endovascular bacteriophage application. *EJVES Vasc Forum.* 2022;56:20–23. <https://doi.org/10.1016/j.ejvsvf.2022.02.004>
 94. Püschel A, Skusa R, Bollensdorf A, Gross J. Local treatment of driveline infection with bacteriophages. *Antibiotics.* 2022;11(10):1310. <https://doi.org/10.3390/antibiotics11101310>
 95. Rubalskii E, Ruemke S, Salmoukas C, Boyle EC, Warnecke G, Tudorache I, et al. Bacteriophage therapy for critical infections related to cardiothoracic surgery. *Antibiotics.* 2020;9(5):232. <https://doi.org/10.3390/antibiotics9050232>
 96. Aslam S, Pretorius V, Lehman SM, Morales S, Schooley RT. Novel bacteriophage therapy for treatment of left ventricular assist device infection. *J Heart Lung Transplant.* 2019;38(4):475–476. <https://doi.org/10.1016/j.healun.2019.01.001>
 97. Gilbey T, Ho J, Cooley LA, Petrovic Fabijan A, Iredell JR. Adjunctive bacteriophage therapy for prosthetic valve endocarditis due to *Staphylococcus aureus*. *Med J Aust.* 2019;211(3):142–143.e1. <https://doi.org/10.5694/mja2.50274>
 98. Chan BK, Turner PE, Kim S, Mojibian HR, Elefteriades JA, Narayan D. Phage treatment of an aortic graft infected with *Pseudomonas aeruginosa*. *Evol Med Public Health.* 2018;2018(1):60–66. <https://doi.org/10.1093/emph/eoy005>
 99. Yang Y, Tan X, Xiong M, Liu Z, Lu S, Ge H, et al. A case report of bacteriophage therapy for the treatment of lung infection due to carbapenem-resistant *Pseudomonas aeruginosa*. *Sci Rep.* 2025;15. <https://doi.org/10.1038/s41598-025-17510-3>
 100. Teney C, Poupelin JC, Briot T, Le Bouar M, Fevre C, Daviet F, et al. Phage therapy in a burn patient colonized with extensively drug-resistant *Pseudomonas aeruginosa* responsible for relapsing ventilator-associated pneumonia and bacteremia. *Viruses.* 2024;16(7):1080. <https://doi.org/10.3390/v16071080>
 101. Köhler T, Luscher A, Falconnet L, Resch G, McBride R, Mai Q-A, et al. Personalized aerosolised bacteriophage treatment of a chronic lung infection due to multidrug-resistant *Pseudomonas aeruginosa*. *Nat Commun.* 2023;14:3629. <https://doi.org/10.1038/s41467-023-39370-z>
 102. Li L, Zhong Q, Zhao Y, Bao J, Liu B, Zhong Z, et al. First-in-human application of double-stranded RNA bacteriophage in the treatment of pulmonary *Pseudomonas aeruginosa* infection. *Microb Biotechnol.* 2023;16(5):862–867. <https://doi.org/10.1111/1751-7915.14217>
 103. Haidar G, Chan BK, Cho ST, Hughes Kramer K, Nordstrom HR, Wallace NR, et al. Phage therapy in a lung transplant recipient with cystic fibrosis infected with multidrug-resistant *Burkholderia multivorans*. *Transpl Infect Dis.* 2023;25:e14041. <https://doi.org/10.1111/tid.14041>
 104. Levêque M, Cassir N, Mathias F, Fevre C, Daviet F, Bermudez J, et al. Refractory *Pseudomonas aeruginosa* bronchopulmonary infection after lung transplantation for common variable immunodeficiency despite maximal treatment including IgM/IgA-enriched immunoglobulins and bacteriophage therapy. *Infect Drug Resist.*

- 2023;16:4265–4271. <https://doi.org/10.2147/IDR.S413900>
105. Li J, Yan B, He B, Li L, Zhou X, Wu N, et al. Development of phage resistance in multidrug-resistant *Klebsiella pneumoniae* is associated with reduced virulence: a case report of a personalised phage therapy. *Clin Microbiol Infect*. 2023;29(12):1601.e1–1601.e7. <https://doi.org/10.1016/j.cmi.2023.08.022>
106. Chen P, Liu Z, Tan X, Wang H, Liang Y, Kong Y, et al. Bacteriophage therapy for empyema caused by carbapenem-resistant *Pseudomonas aeruginosa*. *Biosci Trends*. 2022;16(2):158–162. <https://doi.org/10.5582/bst.2022.01147>
107. Wu N, Dai J, Guo M, Li J, Zhou X, Li F, et al. Pre-optimized phage therapy on secondary *Acinetobacter baumannii* infection in four critical COVID-19 patients. *Emerg Microbes Infect*. 2021;10(1):612–618. <https://doi.org/10.1080/22221751.2021.1902754>
108. Maddocks S, Fabijan AP, Ho J, Lin RCY, Ben Zakour NL, Dugan C, et al. Bacteriophage therapy of ventilator-associated pneumonia and empyema caused by *Pseudomonas aeruginosa*. *Am J Respir Crit Care Med*. 2019;200(9):1179–1181. <https://doi.org/10.1164/rccm.201904-0839LE>
109. Aslam S, Courtwright AM, Koval C, Lehman SM, Morales S, Furr CLL, et al. Early clinical experience of bacteriophage therapy in 3 lung transplant recipients. *Am J Transplant*. 2019;19(9):2631–2639. <https://doi.org/10.1111/ajt.15503>
110. Law N, Logan C, Yung G, Furr CLL, Lehman SM, Morales S, et al. Successful adjunctive use of bacteriophage therapy for treatment of multidrug-resistant *Pseudomonas aeruginosa* infection in a cystic fibrosis patient. *Infection*. 2019;47(4):665–668. <https://doi.org/10.1007/s15010-019-01319-0>
111. Cook JD, Hooey PB, Salazar KC, Clark JR, Jain U, Fernando C, et al. Results of TOR001: an open-label single patient study using targeted bacteriophage therapy for the treatment of chronic urinary tract infection. *Int J Antimicrob Agents*. 2025;66:107613. <https://doi.org/10.1016/j.ijantimicag.2025.107613>
112. Cesta N, Fusco A, Ferretti C, Materazzi A, Altieri A, D'Agostini C, et al. Bacteriophage-enhanced doxycycline activity against *Escherichia coli* in chronic bacterial prostatitis. *Int J Antimicrob Agents*. 2025;66:107571. <https://doi.org/10.1016/j.ijantimicag.2025.107571>
113. Johri AV, Johri P, Hoyle N, Nadareishvili L, Pipia L, Nizharadze D. Case report: successful treatment of recurrent *E. coli* infection with bacteriophage therapy for patient suffering from chronic bacterial prostatitis. *Front Pharmacol*. 2023;14:1243824. <https://doi.org/10.3389/fphar.2023.1243824>
114. Le T, Nang SC, Zhao J, Yu HH, Li J, Gill JJ, et al. Therapeutic potential of intravenous phage as standalone therapy for recurrent drug-resistant urinary tract infections. *Antimicrob Agents Chemother*. 2023;67:e00037-23. <https://doi.org/10.1128/aac.00037-23>
115. Johri AV, Johri P, Hoyle N, Pipia L, Nadareishvili L, Nizharadze D. Case report: chronic bacterial prostatitis treated with phage therapy after multiple failed antibiotic treatments. *Front Pharmacol*. 2021;12:692614. <https://doi.org/10.3389/fphar.2021.692614>
116. Rostkowska OM, Międzybrodzki R, Miszkowska-Szyszkowska D, Górski A, Durlik M. Treatment of recurrent urinary tract infections in a 60-year-old kidney transplant recipient: the use of phage therapy. *Transpl Infect Dis*. 2021;23:e13391. <https://doi.org/10.1111/tid.13391>
117. Terwilliger A, Clark J, Karris M, Hernandez-Santos H, Green S, Aslam S, et al. Phage therapy related microbial succession associated with successful clinical outcome for a recurrent urinary tract infection. *Viruses*. 2021;13(10):2049. <https://doi.org/10.3390/v13102049>
118. Bao J, Wu N, Zeng Y, Chen L, Li L, Yang L, et al. Non-active antibiotic and bacteriophage synergism to successfully treat recurrent urinary tract infection caused by extensively drug-resistant *Klebsiella pneumoniae*. *Emerg Microbes Infect*. 2020;9(1):771–774. <https://doi.org/10.1080/22221751.2020.1747950>
119. Kuipers S, Ruth MM, Mientjes M, de Sévaux RGL, van Ingen J. A Dutch case report of successful treatment of chronic relapsing urinary tract infection with bacteriophages in a renal transplant patient. *Antimicrob Agents Chemother*. 2020;64(1):e01281-19. <https://doi.org/10.1128/AAC.01281-19>
120. Khawaldeh A, Morales S, Dillon B, Alavidze Z, Ginn AN, Thomas L, et al. Bacteriophage therapy for refractory *Pseudomonas aeruginosa* urinary tract infection. *J Med Microbiol*. 2011;60(Pt 11):1697–1700. <https://doi.org/10.1099/jmm.0.029744-0>
121. Letkiewicz S, Międzybrodzki R, Fortuna W, Weber-Dąbrowska B, Górski A. Eradication of *Enterococcus faecalis* by phage therapy in chronic bacterial prostatitis—case report. *Folia Microbiol (Praha)*. 2009;54(5):457–461. <https://doi.org/10.1007/s12223-009-0064-z>
122. Costerton JW, Stewart PS, Greenberg EP. Bacterial biofilms: a common cause of persistent infections. *Science*. 1999;284(5418):1318–1322. <https://doi.org/10.1126/science.284.5418.1318>
123. Zimmerli W, Sendi P. Orthopaedic biofilm infections. *APMIS*. 2017;125(4):353–364. <https://doi.org/10.1111/apm.12687>
124. Arciola CR, Campoccia D, Ehrlich GD, Montanaro L. Biofilm-based implant infections in orthopaedics. *Adv*

- Exp Med Biol.* 2015;830:29–46. https://doi.org/10.1007/978-3-319-11038-7_2
125. Patton JS, Byron PR. Inhaling medicines: delivering drugs to the body through the lungs. *Nat Rev Drug Discov.* 2007;6(1):67–74. <https://doi.org/10.1038/nrd2153>
126. Abedon S, Thomas-Abedon C. Phage therapy pharmacology. *Curr Pharm Biotechnol.* 2010;11(1):28–47. <https://doi.org/10.2174/138920110790725410>
127. Ibrahim R, Aranjani JM, Kalikot Valappil V, Nair G. Unveiling the potential bacteriophage therapy: a systematic review. *Future Sci OA.* 2025;11:2468114. <https://doi.org/10.1080/20565623.2025.2468114>
128. Wang H, Yang Y, Xu Y, Chen Y, Zhang W, Liu T, et al. Phage-based delivery systems: engineering, applications, and challenges in nanomedicines. *J Nanobiotechnol.* 2024;22:365. <https://doi.org/10.1186/s12951-024-02576-4>
129. Abedon ST, Danis-Wlodarczyk KM, Wozniak DJ. Phage cocktail development for bacteriophage therapy: toward improving spectrum of activity breadth and depth. *Pharmaceuticals.* 2021;14(10):1019. <https://doi.org/10.3390/ph14101019>
130. Pirnay J-P, De Vos D, Verbeken G, Merabishvili M, Chianishvili N, Vaneechoutte M, et al. The phage therapy paradigm: prêt-à-porter or sur-mesure? *Pharm Res.* 2011;28(4):934–937. <https://doi.org/10.1007/s11095-010-0313-5>
131. Aslam S, Lampley E, Wooten D, Karris M, Benson C, Strathdee S, et al. Lessons learned from the first 10 consecutive cases of intravenous bacteriophage therapy to treat multidrug-resistant bacterial infections at a single center in the United States. *Open Forum Infect Dis.* 2020;7(9):ofaa389. <https://doi.org/10.1093/ofid/ofaa389>
132. Verbeken G, Pirnay J-P, Lavigne R, Jennes S, De Vos D, Casteels M, et al. Call for a dedicated European legal framework for bacteriophage therapy. *Arch Immunol Ther Exp (Warsz).* 2014;62(2):117–129. <https://doi.org/10.1007/s00005-014-0269-y>
133. Heyse S, Hanna LF, Woolston J, Sulakvelidze A, Charbonneau D. Bacteriophage cocktail for biocontrol of *Salmonella* in dried pet food. *J Food Prot.* 2015;78(1):97–103. <https://doi.org/10.4315/0362-028X.JFP-14-041>
134. Magnone JP, Marek PJ, Sulakvelidze A, Senecal AG. Additive approach for inactivation of *Escherichia coli* O157:H7, *Salmonella*, and *Shigella* spp. on contaminated fresh fruits and vegetables using bacteriophage cocktail and produce wash. *J Food Prot.* 2013;76(8):1336–1341. <https://doi.org/10.4315/0362-028X.JFP-12-517>
135. Roncolato EC, Campos LB, Pessenda G, Costa e Silva L, Furtado GP, Barbosa JE. Phage display as a novel promising antivenom therapy: a review. *Toxicon.* 2015;93:79–84. <https://doi.org/10.1016/j.toxicon.2014.11.001>
136. Siopi M, Skliros D, Paranos P, Koumasi N, Flemetakis E, Pournaras S, et al. Pharmacokinetics and pharmacodynamics of bacteriophage therapy: a review with a focus on multidrug-resistant Gram-negative bacterial infections. *Clin Microbiol Rev.* 2024;37(4):e00044-24. <https://doi.org/10.1128/cmr.00044-24>
137. Blasco L, Bleriot I, Fernández-Grela P, Paño-Pardo JR, Oteo-Iglesias J, Tomás M. Estudios farmacocinéticos y farmacodinámicos de la fagoterapia. *Farm Hosp.* 2025;49(5):407–412. <https://doi.org/10.1016/j.farma.2025.04.003>
138. Fuerst-Wilmes M, Respondek V, Schramm M, Lilienthal N, Buss K, Duechting A. Regulation of phage therapy medicinal products: developments, challenges, and opportunities. *Front Cell Infect Microbiol.* 2025;15:1631359. <https://doi.org/10.3389/fcimb.2025.1631359>
139. Nang SC, Lin Y-W, Petrovic Fabijan A, Chang RYK, Rao GG, Iredell J, et al. Pharmacokinetics/pharmacodynamics of phage therapy: a major hurdle to clinical translation. *Clin Microbiol Infect.* 2023;29(6):702–709. <https://doi.org/10.1016/j.cmi.2023.01.021>
140. Holger D, Kebriaei R, Morrisette T, Lev K, Alexander J, Rybak M. Clinical pharmacology of bacteriophage therapy: a focus on multidrug-resistant *Pseudomonas aeruginosa* infections. *Antibiotics.* 2021;10(5):556. <https://doi.org/10.3390/antibiotics10050556>
141. Knezevic P, Hoyle NS, Matsuzaki S, Gorski A. Editorial: advances in phage therapy: present challenges and future perspectives. *Front Microbiol.* 2021;12:701898. <https://doi.org/10.3389/fmicb.2021.701898>
142. Pirnay JP. Phage therapy in the year 2035. *Front Microbiol.* 2020;11:1171. <https://doi.org/10.3389/fmicb.2020.01171>
143. INFARMED – Autoridade Nacional do Medicamento e Produtos de Saúde, I.P. Deliberação n.º 112/CD/2024: norma orientadora sobre a utilização de medicamentos manipulados para terapia fágica em contexto hospitalar — preparações magistrais de bacteriófagos. Lisboa: INFARMED; 2024 Nov 15. https://www.infarmed.pt/documents/15786/9697484/2024-11-15_Delibera%C3%A7%C3%A3o_112_CD_2024/d0424242-a103-1256-cd7d-526aaf12dfac?version=1.0
144. University of Minho. UMinho announces creation of Therapeutic Phage Production Laboratory [Internet]. Braga: University of Minho; 2025 Oct 27. <https://www.engium.uminho.pt/en/uminho-announces-creation-of-therapeutic-phage-production-laboratory/>

145. Pires DP, Rita Costa A, Pinto G, Meneses L, Azeredo J. Current challenges and future opportunities of phage therapy. *FEMS Microbiol Rev.* 2020;44(6):684–700. <https://doi.org/10.1093/femsre/fuaa017>
146. Pires DP, Meneses L, Brandão AC, Azeredo J. An overview of the current state of phage therapy for the treatment of biofilm-related infections. *Curr Opin Virol.* 2022;53:101209. <https://doi.org/10.1016/j.coviro.2022.101209>
147. Technophage. New INFARMED guidance on the use of phage therapy in the hospital context [Internet]. Lisbon: Technophage; 2024. <https://technophage.pt/new-infarmed-guidance-on-the-use-of-phage-therapy-in-the-hospital-context/>
148. LxBio. LxBio is the 1st Portuguese biopharmaceutical company to join PhageEU, strengthening our commitment to advancing bacteriophage research and innovation [Internet]. Lisbon: LxBio; 2025 Feb 4. <https://www.lxbio.pt/noticiasprpartnership-1>
149. iMed.Ulisboa. Phage Biology Research and Infection Control (PhaBRIC) Laboratory [Internet]. Lisbon: Instituto de Investigação do Medicamento, Universidade de Lisboa; 2025. <https://imed.ulisboa.pt/labs/phage-biology-research-and-infection-control/>
150. i3S. BeSurf – BioEngineered Surfaces Group [Internet]. Porto: Instituto de Investigação e Inovação em Saúde (i3S), University of Porto; 2025. <https://www.i3s.up.pt/research-group.php?groupid=106>
151. International Iberian Nanotechnology Laboratory (INL). Phages on Chip [Internet]. Braga: International Iberian Nanotechnology Laboratory; 2025. <https://www.inl.int/projects/phages-on-chip/>