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Phage therapy as a modern alternative to antibiotics

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Abstract: The global rise of multidrug-resistant (MDR) bacterial infections has accelerated the pursuit of novel antimicrobial strategies. Bacteriophage therapy, which employs viruses that selectively infect and kill bacteria, has re-emerged as a promising alternative due to its host specificity and minimal disruption to commensal flora. Recent innovations in genetic engineering have enabled the creation of recombinant phages and lytic enzymes, enhancing their clinical efficacy. In particular, phage-derived lysins have demonstrated potent bactericidal effects against MDR pathogens, including MRSA and *Acinetobacter baumannii*, in both in vitro and in vivo models. Furthermore, engineered phages using CRISPR/Cas and synthetic biology are opening new frontiers, allowing for targeted eradication of resistance genes and improved biofilm disruption. These therapies are being explored not only for human infections but also for applications in food safety, veterinary medicine, and medical device sanitation. While host immune interactions and pharmacokinetics remain under investigation, current clinical data support the safety and therapeutic promise of phage-based approaches. This review highlights the biological principles, technological advancements, and practical implications of phage therapy as a modern solution to antibiotic resistance.

Keywords: *Antimicrobial resistance, Lytic enzymes, Phage cocktails, Phage therapy, Therapeutic innovation.*

1. Introduction

The idea that treating a bacterial virus for the bacterial infection is gained traction in response to the recent emergence of multidrug-resistant pathogens, but it has been almost a century. Early observations of phage-induced bacterial lysis, their biological properties as well as their therapeutic value have been controversial. Frederick Twort was the first to describe a characteristic area related to phage infection in 1915, but the origin of this phenomenon was attributed to the bacterial virus and the use of the term "bacteriophage" (literally "bacteria eater") [1]. It was also d'Hérelle who devised the idea of using phage as a cure, and in 1919 Hôpital des Enfants-Malades in Paris reported the first case of clinical use of phage. Phage was successfully used in the treatment of four pediatric cases of bacterial dysentery [2]. Despite its initial experimentation, d'Hérelle has achieved several successful results.And his research was strongly contested in the scientific community [3]. Nonetheless d'Hérelle uses dysentery, cholera, and treatment of infectious diseases in the 20th century, has a series of phage therapy center and a commercial phage production facility in Europe and India, Global continued pioneering phage therapy [4]. 1931, one kind of test of phage therapy for the treatment of cholera in the Punjab region of India has included 118 patients and 73 control subjects who received the group phage therapy. d'Hérelle showed 74 deaths in the control group and only 5 deaths in the experimental group, a mortality rate of 90% [2].

There have been many mistakes in these early attempts to treat phage, and most probably did not understand the biological properties of the phage. The basic purification and storage protocol resulted in low potency of active phage and contamination from bacterial antigens, and phages that were not infectious to the target bacteria were used for treatment [1]. In addition, the transmission of the phage

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to the infected area was embarrassing because of the medical limitations of the time. For example, the role of the innate immune response in patients who have removed active phage and reduced the efficacy of phage therapy has recently been observed as a physiological mechanism [5]. As a result, phage therapy has been widely disseminated by most western medicine since the introduction of pharmaceutical antibiotics in the 1940s. Exceptions are in the former Soviet Union and Eastern Europe where clinical phage therapy has been extensively used to treat antibiotic resistant infections caused by a variety of infectious bacteria such as *Staphylococcus*, *Pseudomonas*, *Klebsiella*, and *E. coli* [6, 7].

1.1. Phage Biology Basics

Phage, a naturally occurring bacterial parasite, cannot replicate independently and is ultimately reliant on its bacterial host for survival. Phages, simple but diverse biological organisms, are composed of DNA or RNA enclosed within a protein capsid. Phages typically bind to specific receptors on the surface of the bacterial cell, inject their genetic material into host cells, followed by integration of this material into the bacterial genome (so-called "temperate" phages) and its propagation vertically into daughter cells, or hijacking a bacterial cloning machine for production of next generation phage progeny and lysed cells (so-called "lytic" phages). When the phage progeny reaches critical mass, where some viral particles may contain more than 1000 virus particles depending on the environmental factors, the lytic protein is activated to hydrolyze the peptidoglycan cell wall to induce the release of new phages for resumption of the solute cycle [8, 9].

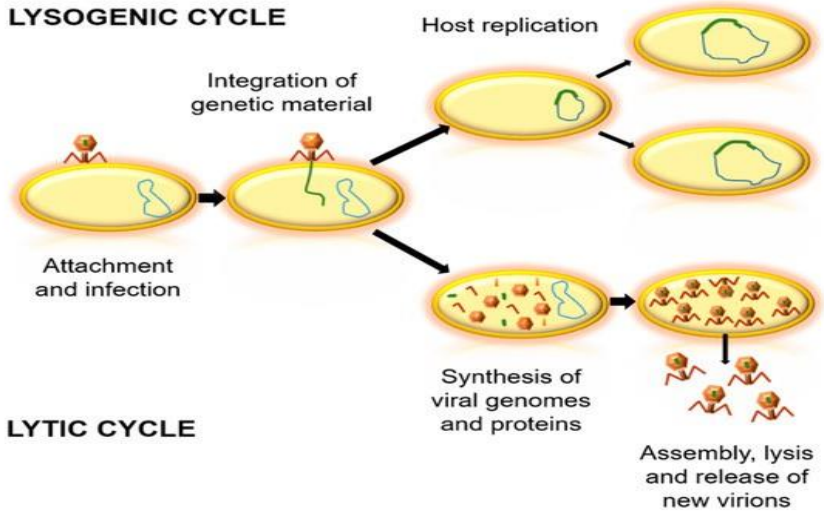

Figure 1.
Biological circulation of phage.

First, the virus binds to bacterial cells, followed by injection of genetic material. In the cycle of viral replication, viral genetic material is integrated into the genome of the host, followed by replication of bacterial cells. During the solute cycle, the viral genetic material is replicated followed by synthesis of viral proteins. A set of virions is then established, followed by the dissolution of bacteria and the release of new virions [10].

1.2. Main Applications of Phage Therapy

As mentioned previously, phage therapy has been utilized for centuries. However, guidelines for conduct of clinical trials and research articles published in Russian or other non-English speaking languages are used only in other eastern countries. Therefore, phage therapy is not currently used in Europe and North American countries. Researchers are currently conducting testing for clinical trials.

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An interesting European project launched by the European Commission in 2013 is currently underway. This project is based on the use of phage cocktails for treatment of burn injuries infected with *Pseudomonas aeruginosa* or *E.coli*.

The primary use of phage therapy is for treatment of diseases and infections as well as for eradication of pathogenic bacteria capable of forming biofilms. Another interesting approach is the use of phage as a preventive disinfectant, particularly in the medical field and with use of clinical instruments. In addition, current technology or a combination of other technologies and phage can be helpful in addressing these clinical applications. Despite the effects of a single type of bacteriophage on bacterial strains due to their high specificity, phage cocktails are considered an interesting strategy for lowering the resistance of bacteria and solving problems related to their range of action [11]. They typically consist of other bacteria capable of attacking other bacterial species or species. In this way, the phage cocktail is able to postpone the emergence of resistance to phage cocktail, thereby playing a decisive role in biofilm formation and the effects of a long-lasting grip. In addition, these cocktails have been proven to offer other benefits including decontaminating food by removing *E. coli*, *Salmonella enterica* or *Listeria* [12].

Phage therapy plays has an important function in other areas, including external medical upcoming food production and livestock rearing. Phage, which can eradicate bacterial infections in animals and prevent ingestion of contaminated food, can be useful in ensuring food safety. One interesting example of the use of phage is for management of a typical food-related infection caused by *Salmonella* (salmonellosis), *Listeria monocytogenes*, *Campylobacter* or *Escherichia coli* (*E. coli*) [13].

1.3. Benefits and Drawbacks of Phage Therapy

Bacteriophage therapy, which can be administered as a treatment for bacterial diseases, offers several benefits, including its potential as a powerful tool in the effort to prevent the emergence of bacterial resistance. One of the main concerns regarding antibiotics is that they can affect microorganisms associated with disease or human imbalance. Because of this connection, the specificity of the phage is based on the fact that replication will occur within a particular host (phage because they cannot infect eukaryotic cells) to resolve the problem. Unlike antibiotics, removal of phages can occur during rapid multiplication in the host, and (if you identify a host only), can be administered in small amounts, with long intervals between, and one population has been removed [14]. Because phage replication can only occur in bacteria, the action of phage inside the host is unusual. Conversely, antibiotics are less accurate and cover more areas if the organism is free of bacteria [15].

Another benefit of phage is that it can be used on areas of the body that are difficult to reach, such as treatment of infections of the central nervous system, which can be a cause of serious problems [3]. An important feature of phage is that it is capable of evolving, and antibiotics are considered static substances that do not change even when the environment changes. Another interesting feature of phage is isolation and production cost. The cost of producing antibiotics is economically high. Antibiotics are not naturally occurring and should be synthesized in the laboratory [16].

The substantial advantage of the specificity of the phage and the limitations of phage therapy is worth noting. Phage should be applied first to minimize potential damage to the microorganism, however, determining which bacteria is causing the disease in vitro is required. This can be a difficult process because rapid identification is required so that the patient can receive treatment [17, 18]. One method of overcoming this obstacle is to use a phage cocktail [14]. However, in vitro and in vivo phage behavior may differ, and in vivo studies are insufficient to ensure an adequate effect [19].

The pharmacology of Phage can be very complex in pharmacokinetics (the action of phages inside the body) and pharmacokinetics (the function of the body on the phages) [20]. In phage therapy, the interaction between phage and bacteria is related to pharmacokinetics. Regarding pharmacokinetics, this is believed to be related to the density of phage in the host. Smaller bacterial populations should require the use of large quantities of phage for more rapid replication when compared with bacteria. In addition, in the case of low bacterial density, replication of the phage will not occur rapidly enough and the

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desired action may not be carried out [21]. This is an indication that there may be variation depending on the dose of phage. The dose of phage is dependent on bacterial density, phage particle size, and phage toxicity. The more toxic the phage is, the more likely it is to attack the host. The solution at this point is based on a toxic phage with a large burst size (producer of large offspring within a short time) and is administered specifically at the site of infection. In addition, there is potential for recognition of the phage or a product by the immune system for induction of an immune response. However, phage dissolution generally occurs more rapidly when compared with retention of neutralizing antibody. However, some researchers have suggested that the potential for an immune response may be related to release of the product and enzymes from the bacterial lysis. According to a recent study, the phage T4 it is highly immunogenic and can be used as potential vaccine candidates [21]. Also, in many cases, the immune response can be evaded by modifying the phage administration mode [20]. Knowledge regarding the immune mechanism involved in detection along with continuous action on the phage is limited. Therefore, an evaluation of the effect of the phage on the human body by examining these substances is needed. By contrast, another study reported that the application of phage therapy had no direct effect on the patient [22]. Despite the fact that the safety of a phage must be verified, phage is consumed indirectly whenever there is fermentation of food, breathing, or accidental drinking of seawater. Thus, bacteriophages do not appear to increase the potential risk [23]. In addition to the pathways by which a phage can naturally reach a bacterial host, there is a new strategy for enhancing the longevity of bacteriophages. Liposomes or capsules around the alginate phage will be accompanied by different ions and must therefore be biocompatible. One question to be addressed is the physical limitations of this structure, and the most appropriate method for encapsulating a phage should be determined according to potential future capabilities [24]. Above all, the most urgent issue to address is the lack of basic information related to dosage, dosage form, protocol, and the appropriate application of this therapy in certain cases of each type of phage [14, 17]. The main question is in regard to the difficulty of digging for a phage (because it is a natural entity) and the failure of pharmaceutical companies to accept this treatment [25]. Legal regulations should be established to define the limitations and safe use of phage therapy. Finally, ethical and social acceptance of phage therapies is a major obstacle to the difficulty of believing that viruses are not only dangerous to humans, but also have the potential to cure diseases [10].

1.4. Phage Against Clinically Significant Pathogens

Recent studies have examined the potential for application of phage therapy for management of clinically important pathogens using animal models. In the treatment of sepsis caused by *P. aeruginosa*, oral administration of phage resulted in a mortality rate of 0% in the experimental group and 66.7% in control mice [26]. A single phage at the same time as administration *of C. difficile* in a hamster model of ileocecitis by *Clostridium difficile* (*C. difficile*) indicated sufficient precautions for infection. After treatment with phage, animals treated with *C. difficile* and clindamycin and control animals died within 9 hours and 11 out of 12 animals in the experimental group survived [27]. The phage combination also caused significantly reduced in vitro growth of *C. difficile*, with limited growth in vivo using the hemostar model [28]. Intraperitoneal administration of a single phage was sufficient to result in 100% rescue mice in a bacteremic model using vancomycin resistant *E. faecium [29] E. coli [30]* producing an expanded β-lactamase, and imipenem resistant *P. aeruginosa [31]*. Promising results have also been obtained using phage cocktails for treatment similar to that for the skin, lungs, and antibiotic-resistant infection of the gastrointestinal tract with *P. aeruginosa* in animal models [26, 32]. Animal experiments using multidrug-resistant *E. coli* O25: H4-ST131 [33], *Vibrio parahaemolyticus [34], S. aureus [35]*, and *A. baumanii* have also been reported. As in the case of multidrug-resistant *P. aeruginosa*, evidence indicating that antibiotic-resistant bacteria can restore antibiotic susceptibility have also been reported [36].

Human experiments in the field of phage therapy have been conducted for nearly a century in various institutions in Eastern Europe, most notably the Eliava Institute for Immunology and

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Experimental Therapy in Wroclaw, Poland. *E. coli*, *S. aureus*, *Streptococcus spp*., *P. aeruginosa*, *Proteus spp*., *Salmonella spp*., *S. dysenteriae* and *Enterococcus spp*., have been used extensively in preclinical and clinical treatments of bacterial pathogens [37]. Effective applications range from surgical treatment to gastrointestinal treatment and prophylactic treatment. In a series of six patients with antibiotic-resistant diabetic ulcers of the foot, topical application of *S. aureus*-specific phage resulted in recovery for all individuals [38]. In a clinical trial conducted in 1938, bacterial dysentery (219 children, 138 children, 81 adults) was used only with phage cocktails composed of various phages, targeting *Shigella flexneri*, *Shigella shiga*, *E. coli*, *Proteus spp*., and treatment with *P. aeruginosa* I was administered. *Salmonella typhi*, *Salmonella paratyphi A* and *B*, *Staphylococcus spp*., *Streptococcus spp*., and *Enterococcus spp*.; cocktails were administered in both oral and rectal form [28]. Of the patients with a history of typhoid fever during the 1974 typhoid epidemic, 28% of the patients who complained of this symptom showed a 27% improvement within 2-3 days [28]. A total of 18,577 children were participants in an interventional trial, which resulted in a 5-fold reduction in the incidence of typhoid fever compared with placebo, with the administration of phage indicating a more effective status for target pathogens when combined with antibiotics [18]. The potential for page treatment remains.

There are currently several commercial page arrangements used for biocontrol of bacterial pathogens that do not have approved paging treatment products for human use in the European Union or the United States but are approved by the food industry as "typically considered safe" by the FDA. In 2011, approximately 48 million cases of food poisoning were reported in the United States alone. Available evidence has suggested that page biocontrol can be an effective method for enhancing food safety at various stages of meat production and processing and also reduce bacterial contamination in fruits, vegetables, and dairy products. Recent placebo controlled human experiments, which demonstrated the safety of page biocontrol and oral page management in these types of food products, are gradually beginning to fill in the knowledge gap regarding the safety of page therapy. These preparations are used mainly for *salmonella spp*, *Listeria monocytase*, MRSA, *E. coli* O157:H7, *Mycobacterium tuberculosis*, *Campylobacter spp*, and *Pseudomonas syringae* [39-42]. There is also potential for detection of pathogens, an example of which is detection of bacillus anthracite using the violin reporter phage [42]. There is increasing evidence of continued enhancement of phase safety with further randomization, double blind and placebo-controlled phase I/II clinical trials of phase therapy, including the establishment of safety and efficacy simultaneously in treating chronic beards caused by antibiotic-resistant *P. aeruginosa* [43]. The innovation of gene editing tools CRISPR / Cas has led to creation of new opportunities for development of phage therapies. One example includes the use of a CRISPR / Cas for destruction of the antibiotic resistance gene for destruction of the antibiotic resistance plasmid using a biotechnology phage [44]. The integration of CRISPR/Cas systems and synthetic biology has paved the way for next-generation phage therapeutics. Bioengineered phages now demonstrate improved biofilm disruption, genetic targeting precision, and in vivo stability, collectively enhancing their clinical utility [45].

1.5. Application of Phage-Derived Lytic Proteins

While the bacterial host is melting, most phage species utilize two major classes of proteins. One is Transembrane protein Holin and the other is endolysisin (lysin), a peptide known as cell wall hydrolase. These two proteins work together to cause the destruction of bacterial cells [46]. Although phage lysine can hydrolyze the bacterial cell wall alone, it cannot dissolve it. Therefore, lysine has received attention as a potential antibacterial agent. This protein can exert a powerful and rapid action and is inert to eukaryotic cells.

Lysins have been obtained successfully from mice in bacteremic strains including multidrug resistant *A. baumannii Streptococcus pneumoniae* [47] and MRSA [48]. Combined use of phage lysine and antibiotics may be more effective in eradicating the infection than in vitro and in vitro proven antibiotics alone in colon models using *C. difficile [49]*. Phage lysin can cause destruction of plant cells, including *B. anthracis* lysin PlyG, which can attack spores in bacillus, a distinct advantage over antibiotics [50].

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Lycin can also be mass produced using common recombinant techniques. Genes for cysteine, histidine - dependent amiodo hydrolase/peptidase (CHAPK), derived from bacteriophage, were cloned and inserted into coliforms for purification. CHAPK Lycin is effective for management of MRSA and it can cause dispersal of S. aureus biofilms [51].

Adding to the appeal of lysins as antibacterial agents, it is considered unlikely that bacteria will develop resistance to lysins due to the fact that they target sites on the peptidoglycan cell wall that are critical for bacterial viability [46]. Operation recombinant phage lysis proteins are much easier to grip due to the short life span, mass production, and administration compared with the actual production of phage, which may be limited by the potential for removal and neutralization of antibodies produced by the host cells of the reticuloendothelial [5]. Future potential for application of phage lysine includes the combination of lysine with the antibiotics considered more effective than antibiotics alone or lysine for treatment of pathogens such as MRSA and C. difficile in mice [52-54]. As shown in Table 1, numerous lytic enzymes derived from bacteriophages have demonstrated potent antibacterial activity against multidrug-resistant (MDR) pathogens such as *Acinetobacter baumannii*, MRSA, and *Pseudomonas aeruginosa*, demonstrated in both in vitro and in vivo models. These findings support the therapeutic promise of phage-derived enzymes in addressing persistent infections

Table 1.
Recently published findings on phage lytic enzymes.

Lysin Type	Enzyme Name	Model System	Target Pathogens
Natural Phage Lysins	ABgp46	In vitro	MDR A. baumannii, P. aeruginosa, S. typhimurium
	PlyF307	Murine	MDR A. baumannii
	Cpl-1	Murine	Streptococcus pneumoniae
	Cocktail (6 lysins)	In vitro, murine in vivo	MRSA
	PlyCD	In vitro, ex vivo	Clostridium difficile
	PlySs2	Murine	S. pyogenes, MRSA
	PlyG	In vitro	Bacillus anthracis
Lysin + Antibiotic	CF-301	Murine	MRSA
	MR-10	Murine	Burn wound infection
Chimeric Lysins	CHAPK	In vitro	MRSA
	ClyH	Murine	MRSA
	Cpl-711	Murine	S. pneumoniae
	Ply187	Murine	Staphylococcus aureus
	Artilysins	Nematode gut, keratinocytes	P. aeruginosa, A. baumannii

Source: Derek, et al. [1].

Additionally, recombinant lysins are being engineered to optimize their therapeutic profile in terms of target specificity, molecular stability, and broader application [55].

1.6. Phage Therapy vs Antibiotic Therapy

Antibiotics and phages both acts as antimicrobial agents capable of breaking down bacterial colonies through dissolution or inhibition, however, there are some differences between antimicrobial agents depending on the situation.

Safety The side effects of antibiotics have been well documented and include numerous gastrointestinal and blood academic complications, including anaphylaxis, renal toxicity, cardiac toxicity, liver toxicity, and neurotoxicity. Most are considered side effects. In such rare cases anaphylaxis is considered a product of a high tissue concentration, related to certain types of antibiotics [56-58]. Contrary to the comprehensive human literature on antibiotic safety, pazy therapy has recently gained attention in Western medicine, and there is currently a significant amount of information on parchment safety. Although oral parchment is generally considered safe [59-62] a major consideration for phage therapy is translocation of the phage across the intestinal epithelium, which is then circulated in the blood [63]. Some data have indicated that translocation of phage through interleukin-2,

inhibition of tumor necrosis factor, and generation of interferon gamma can be beneficial to the host by causing down-regulation of the immune response to the microbial antigen related to the intestinal-response [63]. Other studies have reported that host innate immune responses resulted in elimination of phage following administration in rats [5]. While phage therapists outweigh the shortcomings of nonimmune deficient patients, the immune response to phage can be suggestive of a potential side effect in patients with immunodeficiency, which could lead to hypothetical worsening of the patient's condition. Thus far, other researchers have failed to agree on this possibility, maintaining that phage therapy is unlikely to cause such side effects in immunodeficient patients [37]. Additional complications include the potential for failure of the intestinal barrier resulting from treatment with a phage cocktail. A mouse model was used to demonstrate that oral administration of a commercial Russian-made paces cocktail could increase the levels of plasma in inflammatory circulating immune complexes in the blood and increase intestinal permeability [64]. It could be related to a variety of pathologies. However, another study did not report a significant increase in the level of cytokine in response to phage treatment [65]. The potential of phage therapy, which can interfere with normal barrier function, can seriously affect recent disorders related to disorders of the intestinal barrier, such as Crohn's disease, inflammatory bowel disease [64]. Pincus et al. reported that the inflammatory response to the phage was dependent on the affected area [66]. Obviously, many safety problems related to the treatment of phage still need to be resolved. Physiological responses to the phage can vary based on the individual and may vary depending on the specific phage variants used [1]. Because much of the current research on immunological response to phage is confined to animal models, conduct of future research should focus on clinical testing that includes humans to determine the safety of phage treatment in relation to human health.

Specificity Phage Tends to be specific to species and strains, in contrast to antibiotics. In some cases, it can offer a substantial advantage considering that a wide range of antibiotics have been well documented and incidental to the symbiotic intestinal microflora that are known for causing secondary consequences such as antibiotic-associated diarrhea and C. difficile infection. Other consequences of antibiotic perturbation in the microbial community of the digestive tract include the risk of asthma, obesity, and diabetes [67-69]. Although current understanding of the side effects of phage therapy is limited, compared to antibiotics, phage therapy is less prone to gastrointestinal microbial disruption, while effectively reducing intestinal transport of pathogens including Shigella sonnei and uropathogenic Escherichia coli [70, 71].

Biofilm penetration Despite the proven effectiveness of antibiotics against plankton bacteria including vibrio cholera and the Yersinia plague, so far there have been limits to the treatment of bio-membrane-based bacterial infections [72]. However, an enzyme (e.g. EPS) located outside the capsid of the phage causes the breakdown of extracellular polymeric substances (EPS) and dispersion of bacterial biological membranes to allow the phage to approach bacteria within the EPS matrix [58]. Upon completion of the solute cycle, the exudated offspring of the phage will propagate the dispersion of the biofilm responsible for removal of bacteria from the biological membrane from the subsequent layer [58]. Monitoring is required to determine whether or not antibiotics can inhibit the growth of bacteria when injecting a large amount of high-density biofilm, however, complete eradication by antibiotics is rare, and re-growth will begin to occur after treatment [73, 74]. Although many low-concentration antibiotics are generally regarded as non-toxic, high concentrations can be a cause of tissue toxicity Abedon [58]. Gabisoniya, et al. [75] of the Eliava Institute of Bacteriophages in Tbilisi, Georgia reported that the application of phage to in vitro colonies of the pathogen P. aeruginosa not only prevented additional formation of biofilm by pathogens, but also destroyed existing biofilms. Phage treatment prevented formation of biofilms by L. monocytogenes, P. aeruginosa, and Staphylococcus epidermidis from the surface of medical devices [76]. These results are highly relevant to problems related to infection that persist due to the presence of implantable medical devices, such as catheters, lenses, and prostheses, where biofilm formation is common [1].

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Phage cocktails Due to the tremendous diversity of environmental phages, designing phage cocktails is more complex than the design of combination therapies that include antibiotics. The composition of the phage cocktail is important to ensuring the success of phage therapy. One of the pathological challenges when designing a phage cocktail is whether treatment should include the administration of a standardized cocktail or a customized cocktail. Cocktail design should also consider the grip life cycle. Lysozyme phage appears to be common in indigenous intestinal microorganisms, and prophage accounts for most of the gut virome [77]. Some therapeutically promising solute phages can effectively silence virulence genes in pathogenic bacteria or provide genes for metabolism of short-chain fatty acids, while other lysogenetic phages complement genes for toxicity and antibiotic resistance [78, 79]. This phage cocktail with an intelligent design consisted of four phages not capable of dissolving the A. baumannii host and one phage capable of inhibiting growth only in vitro. Growth inhibition phages were selected for capsular loss against encapsulated A. baumannii. Eliminating the known toxic factor, the capsule can reduce bacterial virulence and is prone to dissolution in four additional phages [80]. This type of cocktail design represents the start of a new treatment option for eradicating bacterial infections that show resistance to conventional therapies. Lysogenic phages have many interesting properties that may be useful in the on-site manipulation of an intracellular microbiome metagenome in an individual bacterium, potentially a human, however, knowledge of the role of lysogenic phage in the human intestine is needed first [81].

2. Conclusion

A major concern regarding antibiotics is that is that they can affect microorganisms associated with disease or human imbalance. Due to this connection, the phage is specific in that replication will occur within a particular host (phage because they cannot infect eukaryotic cells) thereby resolving the problem. Unlike antibiotics, removal of phages can occur when multiplication the host is a rapid occurrence, and (if a host only is found) can be administered in small amounts, with long intervals in between, and one population has been removed [14]. Because phage replication occurs only in bacteria, action of the phage inside the host is unusual. Another benefit of phage is that it can be used on areas of the body that are difficult to reach, such as treatment of infections of the central nervous system, which can lead to serious problems [3]. Although the safety of a phage must be verified, phage is consumed indirectly whenever there is fermenting food, breathing, or accidental drinking of seawater. That is the reason why bacteriophages do not appear to increase the potential risk [23]. Phage lysins have been obtained successfully from mice in bacteremic strains including multidrug resistant *A.baumannii* [82] *Streptococcus pneumoniae* [47] and MRSA [48]. Combined administration of phage lysine and antibiotics may be more effective in eradicating the infection than in vitro and in vitro proven antibiotics alone in colon models using *C. difficile* [49]. Future potential for application of phage lysine includes the combination of lysine with antibiotics considered more effective than antibiotics alone or lysine for treatment of pathogens such as MRSA and *C. difficile* in mice [52-54]. Liposomes, or capsules around the alginate phage, will be accompanied by different ions and must therefore be biocompatible. One question to be addressed is the physical limitations of this structure, and the most appropriate method of encapsulating phage should be determined according to potential capabilities [24]. The appropriate phage cocktail consisted of four phages not capable of dissolving the A. baumannii host and one phage capable of inhibiting growth only in vitro. Growth inhibition phages were selected according to capsular loss against encapsulated *A. baumannii* Eliminating the known toxic factor, the capsule can reduce bacterial virulence and is prone to dissolution in four additional phages [80]. This type of cocktail design represents the establishment of a novel treatment option.

Bacteriophage therapy represents a promising alternative in combating multidrug-resistant bacterial infections. With the development of recombinant lysins, phage cocktails, and CRISPR-based engineering, phage-based strategies continue to evolve toward greater specificity, safety, and therapeutic efficiency. These advancements offer strong potential for application not only in clinical settings but also in food safety, veterinary medicine, and public health. Future studies should focus on

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optimizing delivery systems, addressing immunological responses, and validating large-scale clinical effectiveness.

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