Food Science and Biotechnology (2023) 32:1719–1727
https://doi.org/10.1007/s10068-023-01361-7

INVITED REVIEW

Vibrio-infecting bacteriophages and their potential to control biofilm

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Received: 21 March 2023 / Revised: 8 May 2023 / Accepted: 29 May 2023 / Published online: 19 June 2023

Abstract
The emergence and spread of antibiotic-resistant pathogenic bacteria have necessitated finding new control alternatives. Under these circumstances, lytic bacteriophages offer a viable and promising option. This review focuses on *Vibrio*-infecting bacteriophages and the characteristics that make them suitable for application in the food and aquaculture industries. Bacteria, particularly *Vibrio* spp., can produce biofilms under stress conditions. Therefore, this review summarizes several anti-biofilm mechanisms that phages have, such as stimulating the host bacteria to produce biofilm-degrading enzymes, utilizing tail depolymerases, and penetrating matured biofilms through water channels. Additionally, the advantages of bacteriophages over antibiotics, such as a lower probability of developing resistance and the ability to infect dormant cells, are discussed. Finally, this review presents future research prospects related to further utilization of phages in diverse fields.

Keywords Bacteriophage · *Vibrio* spp. · Antibiotic resistance · Biofilm · Aquaculture

Introduction

Vibrio spp. are Gram-negative, rod-shaped bacteria found in brackish water environments, with their abundance increasing as temperatures rise (Oliver et al., 2018). *Vibrio cholerae* serogroups O1 and O139 cause acute diarrheal disease cholera (CDC, 2022). Among non-cholera vibrios, *Vibrio vulnificus* and *Vibrio parahaemolyticus* are highly pathogenic to humans, with the former having a higher mortality rate (Kim, 2020; Wang et al., 2015). People get infected by these pathogens through consumption of contaminated water/seafood or open wound exposure. By expressing multiple virulence factors, such as the MARTX toxin and phospholipase (Kim et al., 2017; Cho et al., 2022), these non-cholera vibrios often cause severe outcomes including necrotizing fasciitis and septic death. In addition to human-infecting vibrios, there are other species of *Vibrio* (*Vibrio harveyi*, *Vibrio anguillarum* and *Vibrio ordalii*) that can infect marine life, causing a big loss in the aquaculture industry (Plaza et al., 2018).

Based on the Continuous Plankton Recorder survey conducted in the North Atlantic Ocean from 1958 to 2011, it has been reported that the incidence of *Vibrio* spp. is rising due to the increase in sea surface temperature worldwide (Vezzulli et al., 2016). Especially in Korea, the rate of change of sea surface temperature has been reported to be higher than the global average, suggesting that the incidence of *Vibrio*-related diseases is increasing (Korea Meteorological Administration, 2020). In this country, about 10% of the food-borne outbreaks from 2007 to 2012 were attributed to *V. parahaemolyticus*, while the average incidence of *V. vulnificus* infection from 2003 to 2016 was 0.12 cases per 100,000 people but with a case fatality rate as high as 48.9% (Kim et al., 2022; Moon et al., 2014).

Due to the misuse of antibiotics in veterinary medicine, the development of antibiotic-resistant *Vibrio* spp. is an alarming problem. A study conducted in the United States in 2006 revealed that *V. vulnificus* and *V. parahaemolyticus* are resistant to eight or more antibiotics commonly used to treat *Vibrio* infections, as well as ampicillin and chloramphenicol (Elmahdi et al., 2016). Meanwhile, *Vibrio* spp. is a major biofilm forming strain. Nevertheless, antibiotics have shown little effect in controlling biofilm. Bacteria in biofilm

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can transfer resistance genes more easily than those present as plankton cells, increasing the risk of spreading antibiotic resistance. Therefore, new alternatives to target antibiotic-resistant bacteria and/or biofilm need to be developed and deployed (Abe et al., 2020).

A promising solution to this problem is the use of bacteriophages. Bacteriophages are viruses that can only infect and replicate inside their host bacteria. Among these, *Vibrio*-infecting phages are viruses that are ubiquitous in seawater and can only infect bacteria from the *Vibrionaceae* family, being important for the control of the population of *Vibrio* spp. and maintaining the health of the marine ecosystems. These phages have DNA or RNA as their genetic material, are mostly tailed or filamentous and the grand majority follow a lytic infecting cycle. Lytic *Vibrio*-infecting phages are attracting attention as a potential treatment for vibriosis. In addition, they have been proposed as control agents to mitigate antibiotic-resistant and biofilm-forming bacteria, which are currently considered to be significant threats to the food industry (Bischoff et al., 2021).

This review focuses on the use of *Vibrio*-infecting phages as a potential alternative control agent for pathogens causing human infections and losses in aquaculture. Specifically, the current research status of *Vibrio*-infecting phages and their ability to control biofilms will be covered. In addition, the prospects and future opportunities for the use of bacteriophages as biocontrol agents are explored.

Bacteriophages

Phage life cycle

To infect the host bacteria, phages recognize bacterial surface receptors via the receptor binding proteins (RBPs), which are usually located in the phage tail. After this tight adsorption, the phages inject their genome into the host cell. The subsequent infection strategy is determined by whether the phage follows a lytic (virulent) or lysogenic (temperate) life cycle (Salmond and Fineran, 2015). If the phage is a virulent phage, it hijacks the bacterial gene expression machinery to support phage proliferation and assembly of new virions (Madigan et al., 2020). Consequently, the production of phage enzymes, such as holins and endolysins, lyses the bacterial cell wall, allowing the release of new phage progeny and restarting the cycle (Liu et al., 2022b).

Temperate phages, on the other hand, do not replicate inside or kill the host bacteria. Rather, once inside the bacteria, the phage genome is synchronized within the host chromosome as a prophage. Under normal circumstances, the phage genome is kept silent; however, under certain stress conditions, such as exposure to antibiotics, the prophage undergoes an induction cycle that leads to excision of the phage genome and initiation of a lytic cycle (Salmond and Fineran, 2015).

Phages as biocontrol agents

Phages can be used to control pathogenic and multidrug resistant bacteria in a variety of applications, including the food industry. In the food industry, bacteriophages have several advantages over traditional chemical and physical disinfectants. These include effective elimination of target host bacteria, relatively low cost of discovery/production, and minimal impact over the sensory and quality characteristics of the applied food (Moye et al., 2018; O'Sullivan et al., 2019; Sillankorva et al., 2012). In general, in this industry, bacteriophages have been employed to prevent, control, sanitize and preserve food, by applying it on pre-harvested, post-harvested products, food contact surfaces and ready-to-eat foods. With this, a significant reduction of *Salmonella*, *Campylobacter*, *Escherichia coli*, *Listeria monocytogenes* has been achieved (Endersen and Coffey, 2020).

Among the food industries, the aquaculture industry is gradually increasing in size and is considered to play an important role as a future food supplier. However, commercial aquaculture has a high incidence of bacterial diseases, especially vibriosis, due to overfeeding, use of high temperatures, lack of water renewal, and improper removal of injured or dead fish (Almeida et al., 2009). For these problems, phages are likely to be a key solution. In fact, Guenther et al. (2009) demonstrated that when phages were applied to food, there was a greater reduction in food-borne pathogens in the aquatic matrix than in the solid matrix. In addition, Culot et al. (2019) demonstrated that when phages were applied to water, the phages could easily and efficiently enter the body of fish through the gills, facilitating the spread of phages. A number of examples have verified the likelihood of phages to control pathogenic bacteria in aquaculture (Higuera et al., 2013; Karunasagar et al., 2007; Nakai and Park, 2002; Nakai et al., 1999; Park and Nakai, 2003; Park et al., 2000; Silva et al., 2014, 2016; Vinod et al., 2006), proving the efficacy of phages in the aquatic environment. Furthermore, due to their ubiquitous nature, the isolation of bacteriophages is more cost-effective than the development of vaccines or antibiotics (Culot et al., 2019). These characteristics make bacteriophages, especially lytic phages, suitable for use as biocontrol agents in aquaculture.

Currently, there are several phage-related products that have been classified as Generally Recognized as Safe (GRAS) and are commercially available. These products can be used in food products to reduce contamination by some of the major pathogenic and antibiotic resistant bacteria, such as *E. coli* (Ecolicide PX™) (Vikram et al., 2021), *Listeria* (ListShield™) (Lang, 2006), and *Salmonella* (PhageGuard S™) (Parveen et al., 2017; Ye et al., 2022). Currently, there

are no commercially available phage products for the treatment of pathogenic *Vibrio* spp. However, there are multiple ongoing efforts to characterize and evaluate the efficacy of novel bacteriophages that can infect *Vibrio* spp. in food and aquaculture products (Table 1), resulting in several *Vibrio*-infecting products that are under development (Hodgson, 2013; Letchumanan et al., 2016; Richards, 2014).

In the meantime, several studies have been conducted to confirm the safety of phage treatment. According to a review from 2008 to 2021 made by Yang et al. (2022b), the administration of phages (which have various bacteria as hosts, including *V. parahaemolyticus*) to animals and humans did not manifest any major adverse events. In 52 studies investigating various routes of administration, adverse effects were reported in only 7% of the patients treated with phages, and these adverse effects were generally mild and resolvable (Uyttebroek et al., 2022). Based on these reports, the use of phages is considered safe; however, there is still a need to develop an appropriate standard for phage administration.

Biofilm and phages

Vibrio biofilms

Bacteria have developed several mechanisms to withstand external stress. One of these mechanisms is the formation of biofilms. Biofilms are a collection of bacterial communities embedded in a self-produced extracellular polymeric substance (EPS) composed of polysaccharides, proteins, lipids, and extracellular DNA that can adhere to surfaces. Cells within a biofilm are protected from various external stresses (e.g., dehydration, starvation, and predation), are tolerant to antimicrobial agents, and can readily adapt to different environments (Azeredo et al., 2021).

In the case of *Vibrio* spp., different loci (*vps, wcr, syp* and *cps*) control the production of different exopolysaccharides, and thus the production of the exopolysaccharide differs depending on the genes within a particular locus present in the bacteria (Yildiz and Visick, 2009). Notably, the expressions of these *Vibrio* exopolysaccharide genes are controlled by quorum sensing, a bacterial communication system. Therefore, species specific quorum sensing system determine biofilm formation in *Vibrio* spp. (Yildiz and Visick, 2009). The flagellum and pili also play an important role in *Vibrio* biofilm formation. Research has shown that when the bacterium loses the flagellar genes *flaA*, *flrC, flgD* or *flgE*, the attachment ability is significantly reduced and only a weak 3-dimensional biofilm is formed. A similar situation was also observed when the mutations occurred in the main pili (MSHA, ChiRP and PilD) (Yildiz and Visick, 2009).

Although there are not as many studies as with other bacterial strains, *Vibrio* infecting phages have been used to disrupt the biofilm structure. Yang et al. (2022b) used phage FE11 to control the biofilm produced by *V. parahaemolyticus*. The results showed that the ability to disrupt the biofilm was directly related to the concentration of the phage, with the highest concentration used being the one that reduced the bacterial load the most. Matamp and Bhat (2020) achieved an 84% reduction of *V. parahaemolyticus* biofilm after 24 h of phage ϕVP-1 treatment. Furthermore, Tan et al. (2015) applied phage ΦH20 to *V. anguillarum* biofilm, which resulted in a reduction of the total biofilm area from 41,000 μm^2 per mm^2 filter to 5000 μm^2 per mm^2 filter after 6 h of incubation. These are promising results suggesting that phages have multiple mechanisms to overcome biofilm production by *Vibrio* spp.

Role of phages in control of biofilms

To overcome the EPS barrier, lytic phages have developed several attack strategies (Fig. 1). The first mechanism involves stimulating the host bacteria to produce EPS-degrading enzymes after lytic infection. Once the EPS-degrading enzymes break down the pieces of the biofilm, phages and phage progeny can easily penetrate deep and maintain the infection cycle (Amankwah et al., 2021). For example, *Bacillus subtilis* produces the hydrolase γ-PGA, which is encoded by the *pghP* gene of phage ΦNIT. This enzyme breaks down the capsular polysaccharide (poly-γ-glutamate) into tri-, tetra-, and penta-γ-glutamate, facilitating phage movement even through thick biofilms (Geredew et al., 2019; Kimura and Itoh, 2003).

Another mechanism that phages can use to degrade the biofilm is to encode tail depolymerases that can digest the polysaccharides, lipids, and proteins that make up the EPS. Matamp and Bhat (2020) discovered that the *Vibrio*-infecting phage ϕVP-1 can produce a tail tube protein that functions as a polysaccharide-hydrolyzing enzyme that aids in biofilm destruction. A study by Latka and Drulis-Kawa (2020) showed that the *K. pneumoniae* infecting phage KP34 produces the depolymerase KP34p57 (homologous to a pectin lyase), an enzyme with antibiofilm capacity, achieving a 60% reduction in biofilm mass after 72 h of treatment with 10^9 PFU/ml. In addition, the bacteriophage JA1 that infects *V. cholerae* O139 produces a lyase capable of depolymerizing the capsular polysaccharide by β-elimination of a 4-substituted uronic acid residue (Linnerborg et al., 2001).

Although some phages do not encode EPS-degrading enzymes, they can diffuse through the water-accessible channels in biofilm and reach the bacterial cells. Water is the most abundant component in biofilms, making up to 97% of the volume in some cases. This property helps to transport nutrients throughout the matrix. Phages can diffuse through

Table 1 List of recently reported *Vibrio* spp.-infecting bacteriophages (From 2020 to 2023)

Phage	Family	Genome size (kb)	Host	Efficacy test	Reference
VVP001	*Siphoviridae*	76.42	*V. vulnificus*	Abalones	(Kim et al., 2021)
vB_VpaP_DE10	*Autographiviridae*	42.87	*V. parahaemolyticus*		(Ye et al., 2022)
VPT02	*Siphoviridae*	120.55	*V. parahaemolyticus*	RTE raw fish flesh slices	(You et al., 2021)
VPG01	*Siphoviridae*	120.02	*V. parahaemolyticus*	Cutting board and seafood item	(Lee et al., 2023)
KIT04	*Demerecviridae*	114.93	*V. parahaemolyticus*		(Tu et al., 2023)
vB_VpP_DE17	*Podoviridae*	43.397	*V. parahaemolyticus*		(Yang et al., 2022a)
F23s1	*Siphoviridae*	76.648	*V. parahaemolyticus*		(Xia et al., 2022)
Vp33	*Podoviridae*		*V. parahaemolyticus*		(Tan et al., 2021a)
Vp22	*Podoviridae*		*V. parahaemolyticus*		(Tan et al., 2021a)
Vp21	*Podoviridae*		*V. parahaemolyticus*		(Tan et al., 2021a)
Vp02	*Podoviridae*		*V. parahaemolyticus*		(Tan et al., 2021a)
Vp08	*Siphoviridae*		*V. parahaemolyticus*		(Tan et al., 2021a)
Vp11	*Siphoviridae*		*V. parahaemolyticus*		(Tan et al., 2021a)
vB_VpaS_PG07	*Siphoviridae*	112.106	*V. parahaemolyticus*		(Ding et al., 2020)
phiTY18	*Myoviridae*	191.5	*V. parahaemolyticus*		(Liu et al., 2022a)
vB_VpP_WS1	*Microviridae*	5.564	*V. parahaemolyticus*		(Xu et al., 2022)
V5	*Inoviridae*	6.658	*V. parahaemolyticus*	Shrimp	(Dubey et al., 2021; Tyagi et al., 2022)
vB_VpS_BA3	*Siphoviridae*	58.648	*V. parahaemolyticus*		(Yang et al., 2020)
vB_VpS_CA8	*Siphoviridae*	58.48	*V. parahaemolyticus*		(Yang et al., 2020)
vB_VpaP_FE11	*Podoviridae*	43.397	*V. parahaemolyticus*		(Yang et al., 2022b)
27Ua.3	*Siphoviridae*	76.890	*V. parahaemolyticus*		(Stoos et al., 2022)
29Fa.3	*Siphoviridae*	79.348	*V. parahaemolyticus*		(Stoos et al., 2022)
31Fb.4	*Siphoviridae*	77.620	*V. parahaemolyticus*		(Stoos et al., 2022)
33Fb.4	*Siphoviridae*	77.632	*V. parahaemolyticus*		(Stoos et al., 2022)
vB_VpaM_PG19	*Microviridae*	5.572	*V. parahaemolyticus*		(Guo et al., 2022)
vB_VpaP_CHI	*Queuovirinae*	57.805	*V. parahaemolyticus*		(Orozco-Ochoa et al., 2023)
vB_VpaP_ALK	*Queuovirinae*	57.805	*V. parahaemolyticus*		(Orozco-Ochoa et al., 2023)
vB_VpaP_M3	*Autographiviridae*	43.446	*V. parahaemolyticus*		(Orozco-Ochoa et al., 2023)
vB_VpaP_C2	*Autographiviridae*	43.494	*V. parahaemolyticus*		(Orozco-Ochoa et al., 2023)
vB_VpaP_M9	*Autographiviridae*	43.268	*V. parahaemolyticus*		(Orozco-Ochoa et al., 2023)
vB_VpaP_M83	*Autographiviridae*	43.901	*V. parahaemolyticus*		(Orozco-Ochoa et al., 2023)
vB_VpaP_MGD2	*Autographiviridae*	45.105	*V. parahaemolyticus*		(Cao et al., 2021)
ϕVP-1	*Myoviridae*	150.764	*V. parahaemolyticus*		(Matamp and Bhat, 2020)
vB_VpaP_AL-1	*Autographiviridae*	42.854	*V. parahaemolyticus*		(González-Gómez et al., 2022)
vB_VpaS_AL-2	*Siphoviridae*	58.457	*V. parahaemolyticus*		(González-Gómez et al., 2022)
vB_VpS_PG28	*Siphoviridae*	82.712	*V. parahaemolyticus*		(Tian et al., 2022)
KIT05	*Podoviridae*	50.628	*V. parahaemolyticus*		(Anh et al., 2022)
vB_VpaP_GHSM17	*Autographiviridae*	43	*V. parahaemolyticus*		(Liang et al., 2022)
vB_VcaS_HC	*Siphoviridae*	81.566	*V. campbellii*		(Li et al., 2021a)
OPA17	*Siphoviridae*	75.897	*V. campbellii*		(Srisangthong et al., 2023)
ΦImVa-1	*Schitoviridae*	77.479	*V. alginolyticus*		(Tajuddin et al., 2022)
Φ-5	*Myoviridae*	238.053	*V. alginolyticus*	Oyster larvae	(Le et al., 2020)
Φ-6	*Myoviridae*		*V. alginolyticus*	Oyster larvae	(Le et al., 2020)
Φ-7	*Myoviridae*		*V. alginolyticus*	Oyster larvae	(Le et al., 2020)
BUCT549	*Siphoviridae*	80.294	*V. alginolyticus*		(Li et al., 2021b)
vB_ValP_VA-RY-3	*Podoviridae*	40.271	*V. alginolyticus*		(Ren et al., 2022)

Table 1 (continued)

Phage	Family	Genome size (kb)	Host	Efficacy test	Reference
VPMCC5	*Zobellviridae*	48.938	*V. harveyi*		(Kar et al., 2022)
vB_VhaM_pir03	*Myoviridae*	286.284	*V. harveyi*	Brine shrimp	(Misol et al., 2020)
V-YDF132	*Siphoviridae*	84.375	*V. harveyi*		(Kang et al., 2022)
OY1	*Autographiviridae*	43.479	*V. mimicus*		(Gao et al., 2022)
vB_VnaS-L3	*Siphoviridae*	39.99	*V. natriegens*		(Li et al., 2022)

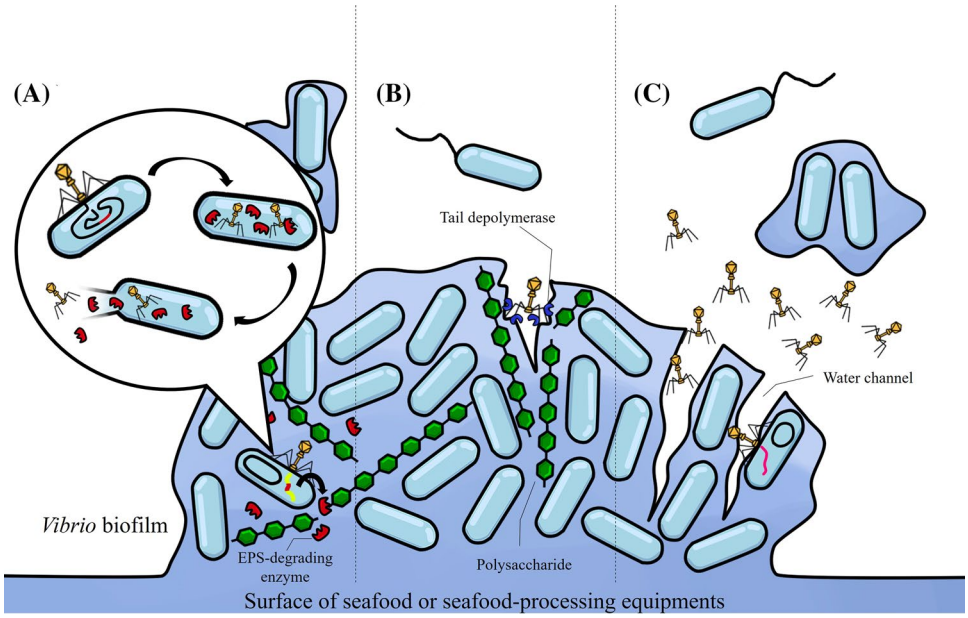

Fig. 1 Biofilm eradication by bacteriophages. (**A**) Phages stimulate the target bacteria to produce enzymes (lyases or hydrolases) that degrade the EPS matrix post phage infection. (**B**) Some phages have enzymes (depolymerases) on their tail spikes that help to degrade the biofilm. (**C**) Water accessible channels in the mature biofilm facilitate the movement of bacteriophages and increase the chance of infection

these water channels and initially infect the bacteria at the edges, penetrating the inner layers of the biofilm by increasing the population through active replication (Azeredo et al., 2021). The results of Vilas Boas et al. (2016) confirmed this observation, as their study showed that during the initial stages of phage infections (specifically using the phage phiIBB-PAA2 with *P. aeruginosa* and vB_AbaP_CEB1 with *Acinetobacter*), the infected cells were primarily located in the outer layer of the biofilm. However, as the infection progressed, cells located at the deepest depths of the biofilm became susceptible to phage infection.

Phages vs. antibiotics to control biofilm in the food industry

One of the main problems with antibiotic treatment is that antibiotics target not only the problematic bacteria, but also the bacteria that make up the normal microbiota, leading to microbiota imbalances. Moreover, due to the slow diffusion of antibiotics within a biofilm, bacteria have a chance to develop resistance mechanisms against the antibiotics, making biofilms difficult to eradicate with such chemicals. In contrast, lytic phages are highly specific and lyse only the target bacteria, likely preserving the normal bacterial flora in the local environment. Although bacteria can also develop resistance mechanisms against phages, phages are living creatures that are constantly evolving and developing new mechanisms to overcome bacterial resistance (Samson et al., 2013). Phages also produce endolysins, enzymes that lyse the peptidoglycan layer, to which bacteria are less likely to develop resistance mechanisms since these enzymes target a highly essential and thus conserved area of the bacterial cell wall (Schmelcher et al., 2012). However, the high specificity of phages to target host bacteria can be a drawback for biofilm treatment, as the biofilm-forming bacteria need to be identified in order to select the appropriate phages. Biofilms are usually composed of different species in nature. Tan et al. (2021b) discovered that *V. parahaemolyticus* and *Shewanella putrefaciens* interact with each other in a synergistic manner to produce a mixed biofilm, increasing the cell viability, EPS content and biomass of the biofilms compared to the corresponding mono-species biofilms. Therefore, to efficiently target the biofilm microbiome, a mixture of different phages should be considered (Sulakvelidze et al., 2001).

The effectiveness of antibiotics depends on the amount supplied. However, sometimes the required amount is

above the minimum toxic concentration for humans, and thus antibiotics cannot be supplied in sufficient quantities to eliminate the biofilm due to health risks. On the other hand, the toxicity of phage-treated food has not yet been clearly analyzed. However, Plaut and Stibitz (2019) stated that if the phages are administered following Good Manufacturing Practices (GMP), the reported adverse effects are none or relatively small. Following this, the toxicity of medical phage-treatment in humans has demonstrated that phages are not toxic even when applied in concentrations as high as 10^{11} PFU/mL to treat a *P. aeruginosa* infection (Suh et al., 2022).

Because biofilms are composed of EPS, which has hydrophobic or hydrophilic properties, the diffusion efficiency of antibiotics is reduced; thus, the delivery of the antibiotic to the bottom of the biofilm is not guaranteed (Azeredo et al., 2021). Nevertheless, there are studies that have shown that phages use the water channels in biofilm to migrate, reach, and infect the innermost cells in a biofilm (Vilas Boas et al., 2016). In addition, phages can self-replicate within the infected bacterial cell, multiplying in large numbers and spreading the infection to the surrounding cells (Azeredo et al., 2021).

When it comes to eliminating persister cells in biofilms, antibiotics are not an option because they can only affect metabolically active cells. Phages also have a limited effect on this type of cells; however, the advantage of phages is that they can infect non-living bacterial cells, remain dormant inside them, and reactivate once the cells become metabolically active (Harper et al., 2014). As bacterial lysis proceeds, the process releases nutrients into the matrix that help restore normal growth in nearby dormant bacterial cells, allowing the phages to infect them. However, one limitation common to both antibiotic and phage treatment of biofilms is that their effectiveness decreases as the thickness, age, and diversity of the biofilm increases (Azeredo et al., 2021).

Future directions

The threat of vibriosis and *Vibri*o biofilms to the food industry is a significant concern, especially under the growing antibiotic resistance problem. Bacteriophages have emerged as a promising alternative to antibiotics, as they possess a high degree of specificity for their host bacteria and thus, are less likely to harm beneficial bacteria. In addition, phages have demonstrated the ability to eradicate biofilm matrices, thereby eliminating pathogens in the process. While it is true that bacteria can develop resistant mechanisms to block phage infection, phages can evolve on their own and counteract this resistance by developing adaptive systems, such as the anti-CRISPR system.

To expand the usage of bacteriophages, however, further investigation is needed to increase their efficiency and/or efficacy. Research related to the use of phage cocktails or the co-treatment of phages with other antibacterial molecules, such as essential oils (bergamot, lemongrass oil, etc.), free-fatty acids (palmitic, stearic acid, etc.), and natural chelating agents (EDTA, nitrilotriacetic acid, etc.), should be explored to widen the range of target pathogens. These components have a synergistic effect, since they can permeabilize the cell membrane, and thus, increase the effectivity of bacteriophages. Phage engineering should also be utilized to increase host range and burst size while reducing the eclipse and latent periods, as these characteristics could improve phage efficiency. Furthermore, phage enzymes (endolysin and depolymerase) could be engineered to enhance their recognition and degradation ability to various substrates, thus increasing their capacity to eradicate diverse bacterial cell walls and multi-species biofilms. Finally, a regulatory framework to standardize the dosage of phages in food products should be developed.

Overall, the evidence suggests that bacteriophages have the potential to be an effective alternative for controlling pathogens and biofilms, particularly with respect to *Vibrio* spp. This presents a promising breakthrough for the challenges faced by the seafood industry.

Acknowledgements This work was supported by the National Research Foundation of Korea funded by the Ministry of Science and ICT (NRF-2020R1F1A1070168 and NRF- 2022R1F1A1074305 to B.S.K.).

Declarations

Conflict of interest There are no conflicts of interest to declare.

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