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Review

Phytopathogenic *Pseudomonas syringae* as a Threat to Agriculture: Perspectives of a Promising Biological Control Using Bacteriophages and Microorganisms

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Abstract: *Pseudomonas syringae* is a Gram-negative bacterium that infects a wide range of plants, causing significant economic losses in agricultural production. The pathogen exhibits a high degree of genetic and phenotypic diversity, which has led to the classification of *P. syringae* strains into different pathovars based on their host range and disease symptoms. Copper-based products have traditionally been used to manage infections in agriculture, but the emergence of copper-resistant strains has become a significant concern. Biological control is a promising strategy to manage *P. syringae*, as it offers an environmentally friendly and sustainable approach to disease management. The review includes an overview of the biology and epidemiology of *P. syringae*, and of the mechanisms of action of various biological control agents, mainly microorganisms (antagonistic bacteria, and fungi) and bacteriophages. Specifically, this review highlights the renewed interest in bacteriophages (bacteria-infecting viruses) due to their advantages over other eco-friendly management methods, thanks to their bactericidal properties and potential to target specific pathogenic bacteria. The potential benefits and limitations of biological control are also examined, along with research directions to optimize the use of this approach for the management of *P. syringae*.

Keywords: *Pseudomonas* spp.; plant pathogen; antimicrobial resistance; biological control

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1. Introduction

Pseudomonas syringae is a phytopathogenic bacterium species that belongs to the class of *Gammaproteobacteria* and causes worldwide diseases in monocots, herbaceous and woody dicots plant species. Thus far, more than 60 pathovars have been identified in this bacterium species, with each pathovar (pv) infecting specific host plants. Strains of most pathovars typically exhibit narrow host ranges, except for pathovar *syringae*, which has a host range of more than 80 plant species, including stone fruits, pome fruits, other woody hosts, crop plants, and grasses [1–3].

During infection, *P. syringae* has two principal interconnected phases of growth: the epiphytic phase, when bacteria live on the surface of plant tissues, usually the above-ground parts, such as leaves, stems, flowers and fruits; and the endophytic phase, when bacteria enter the plant tissue and colonize the intercellular apoplast space. The symptomatology appears only after bacteria enter the plant and multiply in the apoplast during the endophytic phase [4]. Several studies have shown the ability of *P. syringae* to survive in the environment outside their host plants. Thus, several *P. syringae* pathovars have been

isolated both from diseased plants and non-agricultural habitats such as rivers and even snow [5,6]. It has been suggested that this ability to grow in the environment is one of the factors that could explain the evolution of *P. syringae* and the emergence of highly devastating new strains of this plant pathogen [5,7]. In the environment, epiphytic populations, latent infection, overwintering sites on the infected hosts, orchard groundcovers, weeds, and detached plant parts are reservoirs of inoculum for *P. syringae* [7]. The bacteria are dispersed by wind, rain, insects, infested budwood, and infected nursery stocks [8,9].

A variety of symptoms are associated with *P. syringae* pathovar infection (Figure 1). Among the most observed are flower blasts, dead dormant buds, necrotic leaf spots, discolored and or blackened leaf veins and petioles, spots and blisters on fruit, shoot-tip dieback, and stem cankers [8]. The symptomatology may, however, vary depending on the host, the strain of *P. syringae,* and the environmental conditions. For example, in woody trees, the infection of the woody tissue and the formation of cankers can eventually girdle and kill branches, resulting in the loss of fruiting surface and even the tree's death [1,8]. Even if the tree does not die, the impact of erratic disease outbreaks can last for several years due to the time required for infected trees to replace the lost bearing surface [1].

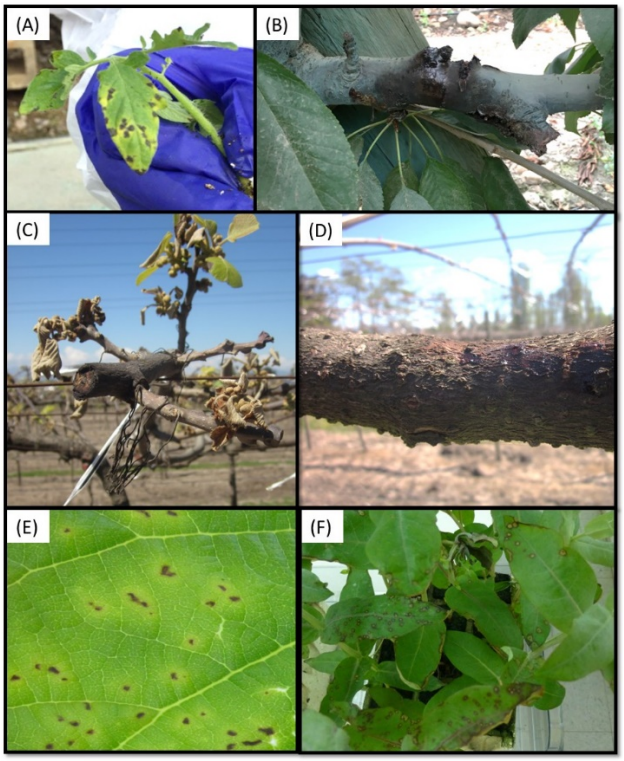

Figure 1. Principal symptoms associated with *Pseudomonas siryngae* infection. (**A**) chlorotic halos on tomato leaves, (**B**) exudate on cherry branch, (**C**) wilting flowers and leaves on kiwi trees, (**D**) cankers with exudate on a kiwi tree branch, (**E**) Kiwi tree leaf with necrotic spots surrounded by a chlorotic halo, and (**F**) necrotic spots on the leaves and the wilting of the vegetative apices in Eucalyptus seedlings (courtesy of Dr. Eugenio Sanfuentes).

Recurrent outbreaks of *P. syringae* pathovars worldwide have stimulated the development of new strategies to contain this pathogen, which is now considered a pandemic that heavily impacts the agriculture industry. Current management of *P. syringae* pathovars consists mainly of chemical and cultural management, but biological control options and synergy between different management strategies have been also recognized as important in the context of a global effort to combat the disease [10,11]. In the case of the chemical management, the use of copper compounds (such as Bordeaux mixture and copper hydroxide), antibiotics (streptomycin and others), and coordination compounds have shown various degrees of success [12]. The principal problem of the above-listed molecules is the

quick development of bacterial resistance, which has limited their usefulness for at least a couple of decades. Many nurseries alternate sprays of copper and streptomycin or combine them to reduce buildup of resistant strains and avoid copper phytotoxicity [11], but this has been insufficient due to the selection of multi-resistant bacterial strains [13,14]. The prevalence of bacteria resistant to agrochemicals has led to concerns due to the accumulation of these compounds in the environment, phytotoxicity, and the shortage of new antibiotics in drug development lines, along with the growing demand for organic or agrochemical-free products. These issues have promoted new management strategies for phytopathogenic bacteria [15,16]. Several studies have suggested developing a sustainable and eco-friendly method for controlling or preventing *P. syringae* pathovar diseases, focusing mainly on biological control. In this review, we will make available the main results obtained in these kinds of study, providing readers with a comprehensive and updated vision of the possible strategies to be implemented for the integrated management of *P. syringae*, together with their possible limitations, in order to establish realistic expectations about the results to be expected, and to clarify the points to address that may be key to achieving future improvement.

According to Eilenberg et al., (2001) [17], biological control is defined as "the use of living organisms (including viruses) to suppress the population density or the impact of a specific pest organism, making it less abundant or less harmful than it would be". In agriculture, biological control mainly consists of the application of non-pathogenic microorganisms to the foliar or root tissues of plants, resulting in the suppression of disease [18]. In this regard, a range of both bacterial and fungal biocontrol agents have been developed and several *P. syringae* pathovars antagonists are commercially available. For example, in the case of *P. syringae* pv. *tomato* (*Pst*), products based in the use of *Bacillus* spp. and *Brevibacillus* bacterial strains are commercially available (baciforte® [19], Nacillus pro® [20] and serenade® max [21]). However, the effectiveness of these biocontrollers is limited due to their incompatibility with antibiotics and copper products. In this scenario, bacteriophages, the viruses of bacteria, have received increased research interest due to their advantages over other management methods, including some that are considered eco-friendly. Bacteriophage-based bactericides can be adapted to target specific disease-causing bacteria [2,22], such as bacterial canker by *P. syringae*. Several studies have reported the isolation and characterization of phages against *P. syringae*, showing their bactericidal properties and potential as biocontrol agents [22–27]. The aim of this review is to analyze and provide updated information about the advances and development of bio control based strategies for the management of the phytopathogen *P. syringae*. This review will be focused on the most relevant biological and pathogenic properties of *P. syringae* worldwide, together with different biological control agents (mainly microorganisms and bacteriophages), highlighting the advantageous characteristics of phage-based methods and the projections for its use as a viable control alternative compatible with a combined management strategy against *P. syringae* pathovars.

2. *Pseudomonas syringae* and Its Damage to Agriculture and Ecosystem

2.1. Identification and Classification of Pseudomonas syringae

Pseudomonas syringae was first reported in 1902 as a pathogenic species of lilac. Since then, *P. syringae* has become recognized as part of a phylogenetic complex (*P. syringae* species complex, Pssc) of ubiquitous strains living in multiple substrates, beyond crops of economic interest [28]. Until a few years ago, the complex included strains with several pathovars of the taxonomically closely related species *P. cichorii* [29], *P. viridiflava* [30], *P. caricapapayae* [31], *P. amygdali* [32], *P. meliae* [33], *P. savastanoi* [34], *P. ficuserectae* [34], *P. avellanae* [35], *P. cannabina* [36], *P. tremae* [36], *P. congelans* [37], *P. asturiensis* [38], *P. cerasi* [39], *P. caspiana* [40], and the not officially recognized *P. coronafaciens* [41]. Historically, the identification and classification of these strains have been based on phenotypic characteristics, such as ecology, physiology, and pathogenicity, which has led to increased taxonomic confusion due to subspecific pathovar names. With DNA–DNA hybridization and multilocus sequence analysis, the genotypic characteristics of these strains allow

a more consistent phylogenetic-based classification [42]. Currently, 62 pathovars have been established in *P. syringae* species based on their pathogenic features [43]. On the other hand, 13 phylogenetic groups (phylogroups) were defined in Pssc by multilocus sequence typing (MLST) on four housekeeping genes; *cts* (encoding citrate synthase), *gapA* (glyceraldehyde-3-phosphate dehydrogenase A), rpoD (RNA polymerase sigma70 factor) and *gyrB* (gyrase B) [44]. Based on comparison of 139 genomes, Gomila et al. (2017) analyzed the taxonomy of Pssc and distinguished 19 phylogenomic species, distributed within 6 phylogenomic branches [42]. Thereby, the Pssc would encompass 11 recognized species: *P. amygdali* (a group denoted by the later synonyms *P. ficuserectae*, *P. meliae*, and *P. savastanoi*), *P. asturiensis*, *P. avellanae*, *P. cannabina*, *P. caricapapayae*, *P. caspiana*, *P. cerasi*, *P. cichorii*, *P. congelans*, *P. syringae*, *P. viridiflava*, along with the supported new *P. coronafaciens* species and representatives of at least 7 putative novel species. Although the Gomila et al. (2017) included 27 strains of *P. syringae* assigned to 15 different pathovars (*aceris*, *actinidiae*, *aptata*, *alisalensis*, *avellanae*, *coriandricola*, *coryli*, *helianthi*, *japonica*, *panici*, *pisi*, *syringae*, *tagetis*, *tomato*, *theae*), the results showed that strains of a particular pathovar (I) clustered together in one phylogenomic species, (II) clustered together with other species strains in one phylogenomic species or (III) are affiliated to different phylogenomic species (Table 1). The study highlights the need to reclassify the misclassified strains and to establish a correct taxonomy for Pssc that can be adopted by the scientific community. These findings are supported by Morris et al. [42], who propose that pathovar denominations do not correspond to the underlying biology of *P. syringae* and are misleading. For Pssc strains identification Guilbaud et al. [43] designed a reliable PCR-based method named *Pseudomonas syringae*-specific polymerase chain reaction (Psy-PCR) that can be used directly with cells from colonies. It demonstrated that 97% of accuracy and sensitivity could be improved with the touchdown method, identifying the 13 phylogroups proposed by Berge et al. [41]. To perform a classification at phylogroup level, partial *cts* gene sequence seems sufficient to accurately predict the phylogenetic affiliation and potential uncertainties can be addressed by comparing additional housekeeping genes [44]. always showing an agreement between Berges's phylogroups and the phylogenomic species proposed by Gomila et al. [39]. In summary, while genomic tools provide a wealth of information that allows us to explore the molecular characteristics of different Pss isolates in depth, in terms of phylogenetic classification, the data obtained via comparison of single or multiple genes are sufficient as a starting point for the classification of Pss isolates.

Table 1. Proposed taxonomy of *Pseudomonas syringae* species complex as in [39] enclosing pathovars distribution.

Phylogenomic Branch	Phylogenomic Species	*P. syringae* Pathovars	Phylogroups [1]
I	*P. congelans* [2]	*syringae*	2c
	P. syringae [2]	*aptata*, *avellanae*, *coryli*, *japonica*, *panici*, *pisi*, *syringae*	2b
	P. cerasi [2]	np	ni
II	Phylogenomic species A [3]	*aceris*, *syringae*	2d
	P. tomato' [3]	*tomato*	1a
	P. avellanae [2]	*actinidae*, *theae*	1b
III	*P. cannabina* [2]	*alisalensis*	5
	P. coriandricola' [3]	*coriandricola*	
	Phylogenomic species B [3]	up	10
IV	*P. coronafaciens'* [3]	np	4
	P. amygdali [2]	np	3
	P. caricapapayae [2]	*helianthi*, *tagetis*	6
V	*P. asturiensis* [2]	np	ni
	Phylogenomic species C [3]	up	9
	P. viridiflava [2]	np	7
VI	*P. cichorii* [2]	np	11
	Phylogenomic species D [3]	up	13
	P. caspiana [2]	np	ni
	Phylogenomic species E [3]	np	ni

[1] According to Berge et al. [41]; [2] Recognized species; [3] Putative novel species; np: no *P. syringae* pathovars in this phylogenomic species; up: *P. syringae* strain with unassigned pathovar; ni: not included in analysis, since not described.

2.2. *Pseudomonas syringae*: A Threat to the Global Agriculture

Disease outbreaks caused by new *P. syringae* isolates continue to threaten global crop production. Some pathovars of *P. syringae* stand out due to their recurrent appearance in several regions of the world. For example, as of 2023, the *syringae* pathovar has been observed in five continents [45], with recent outbreaks in China, Tanzania, Italy, Serbia, Spain, the USA, Iran, Turkey, Australia, Bulgaria, Lithuania, The Netherlands, and the UK, infecting a variety of crops and causing significant economic losses [46,47]. Bacterial canker disease of sweet cherry, tart cherry, apricot, plum, peach and apricot, caused by *P. syringae* pathovars *syringae* and *morsprunorum*, is a concern, and causes economic losses worldwide. For the cherry fruit industry, bacterial canker is an annual problem, being particularly devastating in young orchards, where it can cause the loss of up to 75% of the trees [1,48,49]. Among other worrisome diseases caused by the *syringae* pathovar is the blast and black pit affecting citrus groves such as orange (*Citrus sinensis*) and mandarin (*Citrus rediculate*). This disease occurs in several regions, with an outbreak in Montenegro in 2013 and 2014's spring seasons [50]. *P. syringae* apical necrosis (BAN) of mango (*Mangifera indica*), one of the world's most significant fruit crops, saw an outbreak during 2010–2014 in mango-growing areas of Sicily (southern Italy) [51,52]. *P. syringae* pv *actinidiae* (Psa), starting in 2008, caused severe epidemics in the kiwifruit-growing areas of Asia, Europe, Oceania, and South America, rendering the entire kiwifruit industry vulnerable to the disease [4]. As a result of Psa outbreaks, many countries with solid kiwifruit industries, such as New Zealand, Italy, France, Spain, Portugal, Turkey, Chile, France, Switzerland, Japan, China, and South Korea, have suffered severe economic losses [53–58]. In the case of New Zealand, yield losses caused by the disease in 2012 were estimated to be 21% [46]. The disease caused by Psa was then considered a pandemic, and Psa was placed on the plant quarantine A2 list by the European and Mediterranean Plant Protection Organization [59]. A further example is an isolate of *P. syringae* pv. *aesculin,* which causes bleeding canker disease of horse chestnuts, affecting hundreds of thousands of trees throughout Northwest Europe. A UK-wide survey of horse chestnut trees conducted in 2007 found that over 70% of trees examined had bleeding canker symptoms [46,60]. In annual plants, yields can decrease due to the reduced photosynthetic capacity of infected foliage, defoliation, flower abortion, and fruit lesions, thereby reducing their market value. For example, *P. syringae* pv. *tomato,* the causal agent of bacterial speck in tomato plants, is one of the most devastating pathogens of this crop. An outbreak can cause losses of up to 25% of the seedlings [61], being one of the most destructive aspects of the disease, and the resulting lesions on the fruits' surface make them unsuitable for the market [61,62].

P. syringae pv. *maculicola* (Psm) is becoming a significant pathogen for crucifer crop producers worldwide. Since Psm was described in 1911, many have reported on its diverse phenotypic, genetic, and pathogenic characteristics [63]. In the case of cucurbits plants, a recent outbreak caused by *P. syringae* pv. *lachrymans* was reported in Bangladesh. The plants showed angular leaf spot symptoms, and the disease led to a reduction in cucumber production of up to 37–40% and rendered fruits unmarketable [64]. The disease can also affect several other cultivated and wild cucurbits worldwide, such as watermelon and squash crops, as it was reported in a recent outbreak from southern Georgia to southern Florida, (USA) [65]. Another problematic pathogen is *P. syringae* pv. *pisi* (Ppi) (EPPO A2 quarantine pest), which cause pea (*Pisum sativum*) bacterial blight, a potentially devastating disease that has been described in all pea-producing countries. This pathogen is particularly aggressive in autumn-sown fields in several European countries and Australia, leading to a severe reduction in yield (up to 71%) and seed quality [66]. Generally, in annual plants, disease incidence caused by *P. syringae*, ranged from 50-100% and damage expressed as yield losses could be higher than 25% [47]. The list of concern diseases caused by *P. syringae* strains is very extensive, positioning these bacteria as one of the most studied in agricultural production. Thus, their virulence mechanisms, life cycle and symptoms have been extensively described [47,67,68]. Even so, at present, *P. syringae* pathovars as a whole continue to infect almost all economically important crop species worldwide.

3. Current Methods of *Pseudomonas syringae* Management and Its Principal Limitations

A variety of methods have been tested for management of *P. syringae*, including cultural management (such as proper pruning), use of resistant plants, biological control with microbial antagonists (for example commercially available biological ointments like Nacillus ®), and chemical management based on copper compounds (high solubility copper sulfate pentahydrate, low solubility coppers and Bordeaux), antibiotics (streptomycin, gentamicin and oxytetracycline hydrochloride) and growth regulators, among others [68]. However, the results of these efforts have not always been successful. In fact, the presence and diseases caused by *P. syringae* are still of concern in different crops throughout the world [9,54,69–71]. Regarding the management methods currently used, it has been observed that cultural practices could influence, directly or indirectly, the disease's development, since it is related to permissive environmental conditions such as temperature, humidity, the presence of entry points, and the genetic and physiological features of the host plant, among others [72]. Among cultural management practices we find different training and pruning systems, appropriate fertilization and irrigation rates, controlled soil conditions, cauterization of infected tissues, and the use of selected resistant plants [11,72]. In this sense, a previous study has shown that in the case of Psa, high nitrogen fertilization, iron deficiency, and water stress were related to more severe symptoms of kiwifruit canker [72]. Additionally, in the case of cultural management, other difficulties can be mentioned, for example, pruning tools and mechanical harvesters can transport the pathogen from one plant to another, in addition to causing entry wounds [73]. Furthermore, in the case of Psa, despite the implementation of adequate hygiene measures in growing areas and the cutting and elimination of diseased trees, it has been shown that the spread of the bacteria can continue. This is because the disease can still spread from plants recently infected that have not yet expressed symptoms or in which Psa is present as an epiphyte [73]. The most common treatments against *P. syringae* consist of frequent spraying of orchards with copper derivatives and/or antibiotics, mainly streptomycin (chemical management) [74,75]. Chemical management of *P. syringae* is preventive and is applied at an early stage or in the absence of the disease [73]. However, resistance to copper by *P. syringae* has been described in both fruit trees and annual crops [76,77]. Moreover, the absence of alternatives to copper-based products and their use in high and frequent doses has exacerbated the resistance phenomena [77,78].

The use of antibiotics does not seem to be an effective solution either, since they quickly select for resistance several *P. syringae* strains [73,79,80]. In addition, legal issues must be considered, for example, in Asian countries and New Zealand, the use of streptomycin for the management of plant pathogens is allowed, but this is not the case for Europe, where copper-based compounds are the mainstay of chemical management [74]. Even so, the use of copper remains controversial. In many parts of the world, the use of copper sulfates is no longer recommended as they are highly soluble and toxic to people and the environment. Also, the problem of copper resistant bacteria has led to an increase in the doses necessary to control bacterial diseases in plants, which has triggered the accumulation of copper in soils, causing a negative impact on plants by altering their ability to metabolize nitrogen, root and shoot growth, and induce chlorosis, damaged photosynthetic pigments and sometimes death. Furthermore, human and animal health problems that have been associated with copper toxicity include gastrointestinal, liver, reproductive, and neurodegenerative disorders, such as Alzheimer's disease [80–82]. On the other hand, copper residues can have an effect on the quality parameters of agricultural products, including appearance and/or taste [77].

Unfortunately, both copper and streptomycin present negative environmental and ecological repercussions such as contamination, bioaccumulation, soil pH imbalance, toxicity to the soil biota, problems of phytotoxicity, lack of systemic activity, difficulties to synchronize the applications, bacterial resistance, and residues in the fruit [77,83]. In fact, streptomycin is not a viable management option in many countries, due to the latest research [24]. Furthermore, copper and streptomycin resistance genes have also been

detected in several *P. syringae* strains [80,84]. Other management methods have been studied, for example ethylene bisdithiocarbamate (EBDC), based fungicides as maneb and mancozeb. In this case, contradictory results have been reported by different authors. For example, [85] reported that EBDC-based chemicals alone or in combination with copper -based compounds could be useful for the control of Pst, based on in vitro studies. Although, in the same study, it was reported that Mancozeb (EBDC-based fungicide) alone significantly reduce the disease symptoms in greenhouse conditions but have poor control in the field. In accordance with the above, [86] found that combination of mancozeb (EBDC based fungicide) and copper was ineffective in suppressing foliar and fruit damage caused by Pst in field trials, attributing this ineffectiveness to the presence of copper resistant strains. Thus, these copper-EBDC combinations are only moderately effective and also possibly carcinogenic [87,88].

Recently, plant activators have been introduced and show promising results for controlling bacterial diseases of fruits and vegetables. These are products that stimulate the systemic acquired resistance in plants. Among them, salicylic acid and its functional analogs have shown varying degrees of reduction of the severity of *P. syringae* [89]. For the management of Pst, one of these plant activators is acibenzolar-S-methyl (CGA-245704 or Actigard [Syngenta, Greensboro, NC]). Also, the use of Actigard has been widely adopted as a Psa control measure in the orchard but appears to have variable efficacy on more susceptible plants (as 'Hort16A)' in laboratory and field trials [90]. In this regard, different results have been reported, for example, [91] reported a 50% reduction in Psa disease severity in Actigard-treated 'Hort16A'. In contrast, Michelotti et al. (2018) concluded that Actigard pre-treatment eliminated Psa infection within 48 hours-post-infection resulting in the absence of disease development [92]. However, these first-generation plant activators like Actigard, would not be the final solution, as they can also reduce fruit yield when applied at the recommended regime and suppress effective defense pathways against insect pests [93]. Also, a recent study of Stroud et al., (2022) has concluded that Actigard treatment may have long-term implications for plant health and that Actigard treatment limits Psa population growth *in planta* but fails to eradicate endophytic Psa populations [90].

In the case of Psa, the systemic resistance inducers reported include bacterial proteins such as arpins that activate plant defenses and induce resistance, and polysaccharides such as chitosan. The use of inducers alone is not recommended, and the duration of protection depends on the inducer, pathogen and crop [94]. In recent years, some bioproducts have been developed based on metabolites produced by species of the *Bacillus* genus, which would have control over bacterial canker. However, these should be evaluated within integrated disease management programs [95]. Apparently, the total management of diseases caused by *P. syringae* is practically impossible to achieve, due to the lack of effective management measures and the versatility of this pathogen.

3.1. Antimicrobial Resistance of Pseudomonas syringae

Pseudomonads in general have a reputation for being highly resistant to antimicrobial compounds, and *P. syringae* is no exception. Antimicrobials such as copper and streptomycin have been used for decades to management *P. syringae* infections of crop plants [77,96,97]. The selection of bacterial strains resistant to bactericidal compounds like copper and antibiotics seems to be the main cause of failure in management of pathogens with conventional treatments. Once the resistance genes are acquired, the frequency of resistant strains increases progressively due mainly to horizontal gene transfer between bacteria [98–100]. Below a brief review of the main mechanisms of tolerance to copper and resistance to streptomycin, focusing on those described in *P. syringae* pathovars and strains.

3.1.1. Copper Resistance Mechanisms

In general, microorganisms including bacteria require copper in low concentrations for their metabolic processes. Some bacterial enzymes needed for cell growth and protection against oxidative stress, use copper as cofactor [101]. Normal cellular require-

ments for copper mean that there are a number of copper-chelating proteins naturally expressed in bacteria which are chromosomally controlled. For example, proteins encoded by the *cue*AR operon in *Pseudomonas putida* [102]. Importantly, despite these essential systems using catalytic levels of copper ions, copper can quickly become cytotoxic to bacterial cells, due to the generation of reactive intermediaries which can cause DNA damage, degrade lipids, and disrupt normal protein function leading to cell death [101,102]. The cytotoxicity of copper has been exploited in agricultural systems for bacterial and fungal pathogen management for over 100 years [103]. However, since the mid-1980s there have been increasing reports of copper-tolerance in a wide range of bacterial species important to the agricultural environment, including *Pseudomonas syringae*, *Xanthomonas campestris, Xanthomonas arboricola, Xanthomonas vesicatoria* and *Erwinia amylovora*, among others [77,98,99,104]. In the case of *P. syringae*, copper tolerant strains have been isolated from kiwifruit, apple, tomato, pepper, plum, mango, sweet cherry and snap bean, from cultures in Asia-Pacific, Europe, Africa, North and South America [77].

The mechanisms of resistance to copper have been extensively studied in *P. syringae*. The first described mechanism in *P. syringae* consists of an operon composed of four structural genes *copABCD* (induced by copper) and two regulatory ones encoding the transcription factors *copR* and *copS* [98,105]. The structural *cop* genes are known to encode proteins that are proposed to sense, bind and transport copper ions resulting in the upregulation of the transport and efflux of copper ions, and cell detoxification [98,106]. The study of the genetic determinants of copper resistance has usually revealed the presence of plasmids that carry an operon related to the cop operon of *P. syringae*. In fact, this is the case of copper tolerant strains of Pst, in which the copper-tolerance is generally mediated by a large conjugative plasmid (PT23) carrying *cop* genes [107]. In this context, phylogenetic analysis indicates that individual plasmids of the PT23A family (carrying genetic copper-tolerance determinants) have been transferred between different *P. syringae* pathovars [108,109]. However recent findings suggests that the *copABCD* and *copR/S* operons may be located on either plasmid or chromosomal DNA, depending on the isolate studied [104]. Additional copper-tolerance mechanisms in *P. syringae* continue to be discovered [109,110]. The efflux system *czcCBA* and the analogous system *cusCBA* are members of the heavy metal efflux (HME)-RND (resistance-nodulation-cell division) family. The *czcCBA* system functions in the detoxification of cadmiun, zinc, and cobalt, and the *cusCBA* system works in detoxifying monovalent cations, such as silver and copper. Recently, Gutiérrez-Barranquero et al., 2013 [109] described the presence of a cus system and its relationship with copper resistance in *P. syringae,* and identified a novel arrangement that combine the *copABCD* and *cusCBA* genes along with the *copG* encoded on a conjugative conserved native plasmid, that could be involved in the increase of copper resistance in Pss [109,111]. Also, integrative conjugative elements (ICEs) contributing to copper-tolerance has been identified in Psa strains isolated from New Zealand kiwifruit orchards. In this case, genomic analysis of seven Psa strains, showed that copper resistance, comprising *czc/cusABC* and *copABCD* systems, was acquired via uptake of ICEs, but also plasmids [92].

3.1.2. Streptomycin Resistance Mechanisms

The antibiotic streptomycin has been used in plant disease management since the 1950s and is considered the most effective current chemical treatment against some pathogenic bacteria, including *P. syringae* [55]. However, prolonged use of streptomycin has given rise to streptomycin resistance. Strains of bacterial pathogens as *Erwinia amylovora*, *Erwinia carotovora*, *Xanthomonas campestris*, *P. syringae* pv. *lachrymans*, *P. syringae* pv. *papulans*, Pss, and Psa that are resistant to streptomycin have been isolated in North and South America [55,97,100,112]. Three major mechanisms have been associated with streptomycin resistance: enzymes that modify streptomycin, alterations of the streptomycin ribosomal binding site, and reduced cellular uptake of the antibiotic [113]. Most known streptomycin resistance determinants encode enzymes that confer resistance through inactivation of

the streptomycin molecule through either phosphorylation or adenylation. Generally, the genetic determinant of that kind of resistance consists of tandem *strA-strB* aminoglycoside phosphotransferases genes, which have been found in bacterial plasmids, integrons and transposons, being present in at least 21 bacterial genera including pathogens of humans, animals, and plants [97,112]. For example, in streptomycin resistant strains of Psa isolated from kiwifruit in Japanese orchards, resistance was shown to coincide with the occurrence of one or both two plasmids pPaCu1 and pPaCu2 [55,114]. In some cases, it has been described that a high streptomycin resistance could not be sustained in the presence only of *strA-strB* genes. For example, in a Japanese Psa strain, it was observed that these genes confer only low-level resistance, and the high-level resistance in this strain is probably a result of upstream insertions of sequences acting as strong promoters of streptomycin resistance genes [113]. Another streptomycin inactivating enzyme is the nucleotidyl transferase encoded by the *aadA* gene, which is mainly associated with integrons, facilitating its co-selection with other antibiotic resistance determinants [100,115]. Three other streptomycin-resistance determinants, *aph*(*6*)-*1c*, *ant*(*3″*), and *ant*(*6*), are more limited in distribution at the current time, and have not been largely described for *P. syringae* [97].

As mentioned above, alterations of the streptomycin ribosomal binding site, also results in antibiotic resistance, this is the case of a single point mutation at codon 43 of the *rpsL* gene resulting in a swap of lysine for arginine in the ribosomal protein S12, that has been detected in streptomycin-resistant Psa strains [113]. Additionally, genomic analysis of Psa strains isolated from outbreaks in Japan and Italy, revealed the presence of drug efflux pumps, including genes belonging to four superfamilies of drug efflux transporters: resistance nodular division (RND), major facilitator superfamily (MFS), multidrug endosomal transporter (MET) and multi-antimicrobial resistance (MAR) [116], suggesting its contribution to the development of high-level antibiotic resistance in pathogen strains. Extensive studies on bacterial resistance to antibiotics have revealed a constant appearance of new resistance mechanisms, considering mutations in specific genes that alter the target of the antibiotic; the use of detoxifying enzymes; decreased entry of antibiotics through membrane-associated transporters; and the modification of existing mechanisms, which confers resistance to new antibiotics [117,118]. Additionally, resistance genes are transferred to new hosts, granting multi-resistance in some phytopathogenic bacteria [117,118].

It has been shown that the presence of high concentrations of metals, favor the co-selection of resistance to antibiotics [14], generating co-resistance (presence of different determinants of resistance in the same genetic element) and cross resistance, where the same genetic determinant confers resistance to antibiotics and metals [13]. Currently, there is a growing concern about the co-selection of bacteria resistant to antibiotics and metals (such as copper) due to the excessive use of these compounds in agriculture [13,14]. For example, in the study performed by Vanneste et al. (2008) [119], all the strains of *P. syringae* pathovars found to be resistant to copper were also resistant to streptomycin [119,120]. The situation is aggravated considering the existence of *P. syringae* pathovars strains that carry determinants of resistance to copper and streptomycin in the same plasmid, which could be transferred from one bacterium to another, thus increasing the possibility of selecting strains resistant to one of these compounds when using the other. Consequently, the spread of these resistances in plant pathogenic bacteria would leave very few options for the management of bacterial diseases [119]. Furthermore, in the study of Hwang et al. (2005), from a total of 95 analyzed *P. syringae* pathovars strains, it was observed that most of them were resistant to copper and ampicillin, however, strains simultaneously resistant to chloramphenicol, rifampicin, ampicillin, and copper or to streptomycin and copper, among other combinations, were also observed [96]. In this context, different studies has reported resistant phenotypes of *Pseudomonas* species, including *P. syringae*, associated to agricultural environments, showing that this bacteria not only have resistance to conventional antimicrobials used in agriculture, but also to some used in human health such as ampicillin, chloramphenicol, rifampin, tetracycline, vancomycin and erythromycin, among others [121,122], thus, becoming in an emerging threat to the agriculture industry and to human health.

In this context, some strategies to face the crisis of antibiotic resistance have been developed, based on the implementation of surveillance programs to avoid the abuse in the antibiotics use in agriculture. For example, in America, this is monitored by US Department of Agriculture (USDA) to ensure that the antibiotic residues do not exceed the tolerance levels marked unsafe by FDA (Food and Drug Administration). Also, various other countries such as Sweden, Denmark, India and China have also adopted regulatory measures to some extent [123]. On the other hand, among the scientific community, the principal strategy to overcome antibiotic resistance is the development of alternative compounds or methods to replace the use of antibiotics. Among them, we can find molecular and other scientific methods targeting for example DNA, RNA, proteins, cell wall, cell membrane, an intracellular target, biosynthetic pathways or ribosomes, the use of peptide antibiotics/antimicrobial peptides (AMPs); the use of combinations of antibiotics and other compounds, phenotypic conversion of drug-resistant to drug-sensitive bacteria, and bacteriophage therapy, among others [123–125]. However, except for the use of bacteriophages, these strategies are still difficult to implement in agricultural crops. Currently, the prevalence of bacteria resistant to agrochemicals, the concern about the accumulation of these compounds, and the shortage of new antibiotics in drug development lines, together with the growing demand for organic or agrochemical-free products, have led to efforts to consider and develop new management strategies for plant pathogenic bacteria [15,16]. In this context, biological (mainly microbiological) control agents are considered the most promising and have been the focus of various investigations in recent decades. The next sections will assess the progress in this matter, regarding the management of *P. syringae.*

4. Biological Control of *Pseudomonas syringae*

In the framework of sustainable agriculture, integrated pest management (IPM) is essential. According to Stenberg, (2017) [126], IPM is a holistic strategy to combat diseases and plant pests, like *P. syringae*, using all available methods while minimizing chemical pesticide applications. In this sense, biological control is positioned as an essential strategy within the IPM because it is friendly with the environment compared to chemical management. Figure 2 summarizes the main chemical control strategies currently used and the main biological control agents that have been used (or that have studies that have shown their potential usefulness) for the management of *P. syringae.*

As mentioned before, there are strategies that seek to improve the intrinsic plant resistance to pathogens [127], that might fall within the spectrum of biological control. In line with the strategies that focus on modifying the resistance of the plant, there are several studies in which transgenic plants that acquire diverse levels of resistance to infection by *P. syringae* have been developed [128–131]. In this regard, this strategy has had promising results in experimental model plants, but field studies are still needed; above all, the regulatory aspects of the use of transgenic plants in different countries worldwide should be considered. In this context (and as mentioned above), in this review, we will focus on biological control according to the definition of Eilenberg et al. (2001), mainly considering the role of microorganisms as biological control agents (BCA) [17].

In nature, antagonism is a characteristic interaction of many organisms that interfere due to their position in an ecological niche; when this property is used to manage a pest, it is named BCA. Most organisms can exploit different modes of action in combination or alternation in a sequence of antagonistic events [132]. Among the types of interspecies antagonisms that can lead to biocontrol of plant pathogens are (i) direct antagonism, whose mechanisms are hyperparasitism and predation; (ii) mixed-path antagonism, whose mechanisms correspond to the release of antibiotics, lytic enzymes, unregulated waste products and physical/chemical interference molecules; and (iii) indirect antagonism, whose mechanisms correspond to competition and the induction of host resistance (Table 2) [133]. The following section presents some of the most advanced and promising studies associated

with biological control strategies (fungi, bacteria, and bacteriophages) to manage diseases caused by different pathovars of *P. syringae* that are of economic importance.

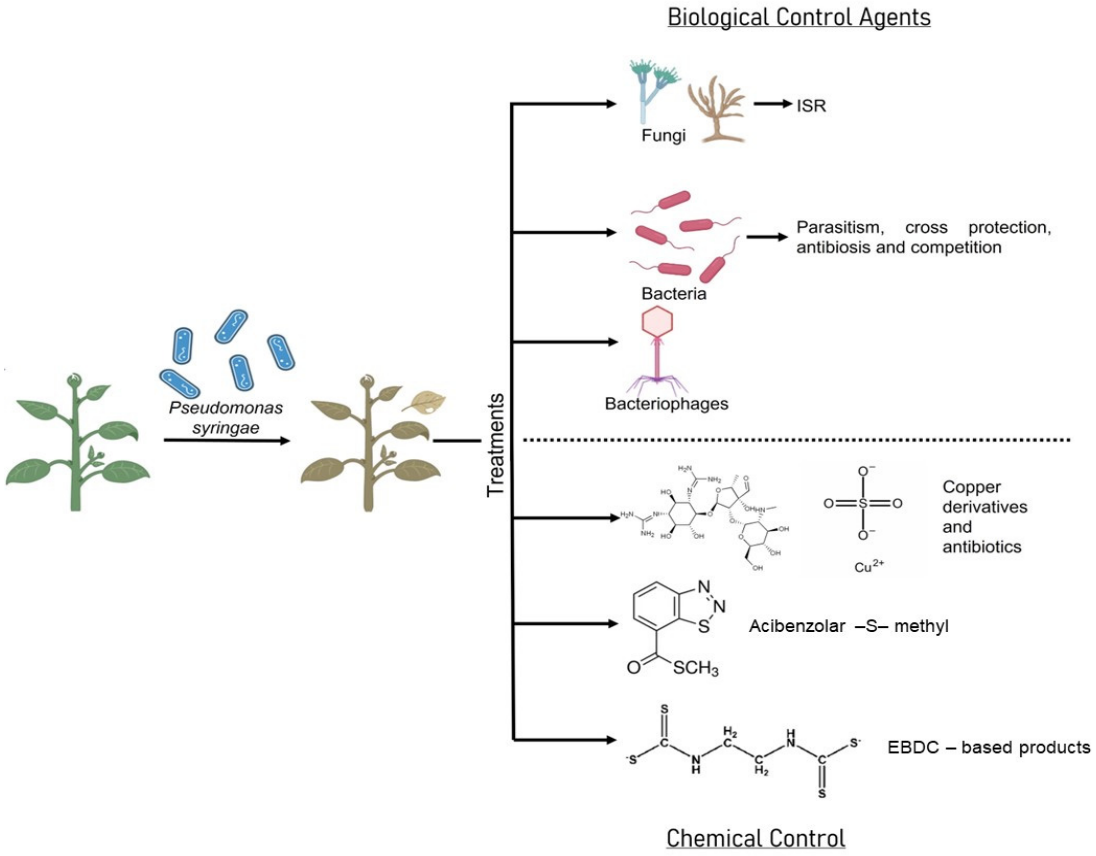

Figure 2. Principal strategies of biological and chemical control against *Pseudomonas syringae*.

Table 2. Progress status of biological control agents studied against *Pseudomonas syringae* pathovars and their interspecies antagonism mechanism.

Type	Mechanism	Biocontrol Agent	Studied *P. syringae* Pathovar [1]	Progress Status [2,3]	References
Direct antagonism	Predation	Bacteriophage	Psa	In vivo studies (1)	[27]
			Psm	In vivo studies (1)	[134]
			Pspo	Field trial (1)	[25]
			Pss	In vivo studies (1)	[134]
			Pst	Phage-based product available (1)	[135]
Mixed-path antagonism	Antibiotics	Bacteria (*Bacillus amyloliquefaciens D747; B. subtilis QST713)*	Psa	Bacteria-based product available (2)	[21,136]
			Pss	In vivo studies (2)	[137,138]
			Pst	Bacteria-based product available (1)	[21]
	Undetermined	Bacteria	Pc	In vitro (1)	[132]
			Pss	Field trial (1)	[139]
Indirect antagonism	Competition	Bacteria *Pseudomonas fluorescens A506*	Pst	Bacteria-based product available (1)	[140]
	Induction of host resistance	Fungi	Psga	In vivo (1)	[122]
			Pst	In vivo (1)	[141]
				In vivo(1)	[132]
				In vivo (1)	[142]

Table 2. *Cont.*

Type	Mechanism	Biocontrol Agent	Studied *P. syringae* Pathovar [1]	Progress Status [2,3]	References
Direct antagonism	Predation	Bacteriophage	Psa	In vivo studies (1)	[27]
			Psm	In vivo studies (1)	[134]
			Pspo	Field trial (1)	[25]
			Pss	In vivo studies (1)	[134]
			Pst	Phage-based product available (1)	[135]
Mixed-path antagonism	Antibiotics	Bacteria *Bacillus amyloliquefaciens D747; B. subtilis QST713*	Psa	Bacteria-based product available (2)	[21,136]
			Pss	In vivo studies (2)	[137]
			Pst	Bacteria-based product available (1) *Bacillus subtilis QST713*	[21]
	Undetermined	Bacteria	Pc	In vitro (1)	[132]
			Pss	Field trial (1)	[139]
Indirect antagonism	Competition	Bacteria *Pseudomonas fluorescens A506*	Pst	Bacteria-based product available (1)	[140]
	Induction of host resistance	Fungi	Psga	In vivo (1)	[122]
			Pst	In vivo (1)	[141]
				In planta (1)	[132]
				In planta (1)	[142]

[1] Psa, P. syringae pv. actinidiae; Pc, P. syringae pv. coryli; Psga, P. syringae pv. garcae; Psm, P. syringae pv. morsprunorum; Pspo, P. syringae pv. porri; Pss, P. syringae pv. syringae; Pst, P. syringae pv. tomato. [2] Numbers in parentheses indicate the number of studies that have reached this level of progress. [3] Field trial: refers to a more advanced experimental phase. Bacteria (or other)-based product available: refers to cases in which a commercially available product has been already developed.

4.1. Fungi

Non-pathogenic fungi, especially plant growth-promoter fungi (PGPF), which are found colonizing plant roots, can act as BCAs and have proven effective against soil-transmitted diseases. They have been recognized for decades for their potential in triggering induced systemic resistance (ISR) in plants through a mechanism that involves biochemical and cytological changes, allowing better management of these diseases [143]. This signaling route permits the plant to generate responses in the affected organs and transmit them to distant organs, as observed with some species of fungi that colonize roots and have effects, via ISR, on aerial organs [144]. In this context, the use of fungi has been studied as a possible BCA for *P. syringae*. In 2007, Hossain et al., [118] showed that the application of both the fungus *Penicillium simplicissimum* GP17-2 and its culture filtrate (CF) generates an ISR in *Arabidopsis thaliana* [141]. Their results showed that *A. thaliana* grown on soil containing GP17-2 can suppress the disease caused by *P. syringae* pv. *tomato*. However, it was revealed that the results were due to a reduction in disease severity rather than a decline in the bacterial population. It is noteworthy that in this case, the reduction of the disease was not caused by contact between both microorganisms, since the authors demonstrated that GP17-2 did not colonize the aerial parts of the plant. By using *A. thaliana* mutant varieties, the authors concluded that genes induced by salicylic acid (SA), jasmonic acid (JA), and ethylene (ET) play an important role in the ISR triggered in plants by GP17-2 or its CF. Based on their results, these researchers suggest that the microorganisms that favor ISR have multiple molecular patterns associated with pathogens (PAMPs), which would be recognized by different receptors, thus activating different signaling pathways in the plant [141]. Botrel et al. evaluated the protective effect of the fungus *Phialomyces macrosporus* against the halo blight caused by *P. syringae* pv. *garcae* (*Psga*) in coffee seedlings [145]. The application of this fungus reduced the disease severity and increased the vegetative growth of seedlings. This saprophytic fungus helps to control the disease through ISR, specifically by increasing the activity of three important enzymes in the plant's defense response: phenylalanine ammonia lyase (PAL), guaiac peroxidase (POX), and ascorbate

peroxidase (APX). The authors demonstrated that this defense response in the plant was not instantaneous and required a period of time to be triggered. According to their results, the foliar application of the fungus *P. macrosporus* to coffee seedlings 7 days before inoculation with Psg induces an increase in the activities of the enzymes mentioned.

Plant ISR can be activated by certain non-pathogenic microorganisms in the rhizosphere through the JA signaling pathway, followed by the ET signaling pathway [141]. In 2018, it was reported that *Aspergillus terreus* was able to suppress the bacterial speck disease caused by *P. syringae* pv. *tomato* DC3000 [142]. Although the mechanism by which the fungus generates the induction of resistance is not clear, it is hypothesized that different metabolites produced by the microorganism could act as inducers of plant defenses [142]. More recently, Köhl et al., (2020) [132] have performed large-scale screenings in the search for microbial antagonists and their derived metabolites against pathogens that affect tomato plants. The authors conducted numerous experiments to measure the rate of disease caused by different pathogens, including *P. syringae* pv. *tomato*, and to test the protective effects of various fungal and bacterial isolates. Among the most effective fungal isolates obtained are *Engyodontium album*, *Verticillium* sp., *Simplicillium lamellicola*, *Guignadia vaccinii*, *Engyodontium parvisporum*, *Lecanicillium tenuipes*, and *Pythium aphanidermatum*. Although the authors demonstrated the promising capacity of these microorganisms in reducing the symptoms caused by Pst, they found no correlation between the use of microorganisms as living cells and the use of the metabolites secreted by them, suggesting that, for a truly beneficial effect, additional studies are required to select metabolites and determine their optimal combination [132]. The use of fungi as biological controllers of phytopathogenic bacteria such as *P. syringae* has several benefits. One of the advantages is the global abundance of fungi, since it is estimated there are still miles (or millions) of species that are not fully known or isolated [146,147]. As a consequence, there is a large repository for direct application on crops, and with which we can carry out massive screenings in search of active compounds that will help us to control diseases. In addition, these microorganisms can activate signaling pathways, thereby inducing a defense response in the plant, which leads to a potentiation of said response. However, this may be difficult in the study of the protective effects of fungi, because both the mutualistic microorganisms and the plant pathogen can induce the signaling pathways involved in the defense mechanisms of the plant. It is still unknown which fungal signals trigger ISR in plants, and fulfilling this knowledge gap will be difficult, considering the complexity of the various signaling pathways involved in the defense responses.

4.2. Bacteria

The use of bacteria as BCAs has been extensively investigated, because genetic and biochemical analyses and the mass production of bacteria (or bacterial products) are much more accessible than fungi. Several BCAs have been reported within the genera *Agrobacterium, Pseudomonas, Bacillus, Alcaligenes,* and *Streptomyces*, among others, and it is expected that this type of biocontroller will have great potential in organic agriculture. Different mechanisms are involved in the protective effects of these microorganisms against plant pathogens (Table 1), such as parasitism, cross-protection, antibiosis, and competition [148]. It is difficult to determine one main mode of action, since a combination of mechanisms usually occurs, and in other cases, these mechanisms have not been elucidated yet. One disadvantage of using bacteria as BCAs is that there are bacterial strains that cannot integrate with copper bactericides and/or antibiotics due to their susceptibility to them. However, there is some potential to integrate these control agents with plant activators (chemicals such as actigard [87] or biologicals such as plant growth-promoting rhizobacteria (PGPR)) [149,150]. They could also be combined with essential oils [137]. When the mechanism of action of the bacterium involves competitive exclusion, the displacement of the pathogenic population from the niche happens, and colonization by the biocontroller bacteria is not certain to occur, which may be ineffective in controlling a

disease. Finally, it could also be that the bacteria used as BCAs release chemical compounds that are phytotoxic or harmful to the host plant microbiota [151].

Several studies have been performed using bacteria as BCAs for *P. syringae*. For example, the antagonist bacteria *Lactobacillus plantarum* PM411 and TC92 were used in field assays to control *P. syringae* pv. *actinidae* (Psa). The results showed that the BCAs prevented the pathogenic bacteria from infecting their host plant. Furthermore, the biocontrolling capacity of *L. plantarum* PM411 and TC92 was comparable to that of the commercial products amylo-X (*Bacillus amyloliquefaciens* D747) and Serenade Max (*B. subtilis* QST713) [152], which have a mode of action associated to the production of lipopeptides that permeabilize the host membrane [153]. Another interesting case of antagonism was reported by Wicaksono et al. (2018) [154], in which Psa infection was inhibited in kiwi trees (*Actinidia deliciosa*), and the disease was attenuated by transferring endophytic bacteria from the shrub *Leptospermum scoparium*, which produces essential oils with antimicrobial properties. The authors suggest that these bacteria are resistant to the antimicrobial compounds present in the vascular system of the host plant, and they could be useful for biocontrol in combination therapies. In the case of *P. syringae* pv. *corilii* (Psc), environmental strains of *Pseudomonas fluorescens* capable of controlling this pathogen in vitro have been described, although the mechanism of action was not elucidated [155]. In this regard, a more recent study identified microbial cytokinin production as a key determinant of the efficient biocontrol effect of *P. fluorescens* against *P. syringae* [156]. On the other hand, mechanisms such as a high production of siderophores and the synthesis of specific secondary metabolites have been reported as key components in the antifungal activity of some *P. fluorescens* strains [157,158]; however, to the best of our knowledge, these mechanisms have not been demonstrated for the biocontrol of *P. syringae*.

Several field studies have been carried out for Pss, with *Pantoea agglomerans* being one of the biocontrollers with the most potential [139]; however, its mechanism of action is still undetermined. *Pantoea* spp. has been extensively studied as a biocontrol agent of different pathogens [159]; previous works have demonstrated that that some *Pantoea* spp. strains are protective through antibiosis activity, or through competition for resources. In addition, application of *Pantoea* spp. to seeds ensures that germinating seedlings are in immediate contact with microbes, and this may prime the plant's immune system so that it is better able to mount a response against pathogens as *P. syringae*, thus indirectly protecting against disease. Results obtained by Morella et al., (2019) suggest that both direct and indirect mechanisms mediating the biocontrol effect of *Pantoea*spp. against *P syringae* [159]. Additionally, results obtained in the study of Akbaba and Ozaktan (2018) suggest that induced systemic resistance (ISR) or nutrient competition may be important factors in the biocontrol of *P. syringae* pv. *lachrymans* (Psl) by an endophytic isolate of *P. agglomerans* [160].

Another example is *Bacillus subtilis* 6051, which controls Pss infection in *A. thaliana* through biofilm formation and the production of surfactin, a cyclic lipopeptide [161]. In an ex vivo study, 206 bacterial strains were screened, of which *Pseudomonas agglomerans* (RK 84, 85, 113 and 154), *Leclercia adecarboxylata* (RK 164), *Pseudomonas putida* (RK 142), *Curtobacterium flaccumfaciens* (RK 114), *Erwinia rhapontici* (RK 135), *Alcaligenes piechaudii* (RK 137) and *Serratia liquefaciens* (RK 102) were found to be potential biocontrollers of Pss [162]. In addition, Mougou and Boughalleb-M'hamdi [115], determined through in vivo assays that 21 *Bacillus* spp. strains were effective in the control of Pss. In this study, combined experiments were also performed using garlic extract, which contains essential oils with controlling potential, thus classified as a green pesticide [163].

Regarding Pst, there are field studies using the commercial product BlightBan A506, whose active component corresponds to the bacterial strain *P. fluorescens* A506, which provided an average of 18% reduction in the disease in nine different field experiments [87], via the mode of action of competitive exclusion. In the same study, the bacterial strains *P. syringae* TLP2 and *P. syringae Cit7*, and *hrp* mutants of Pst DC3000 were used. Treatment with *P. syringae Cit7* was the most effective, providing an average level of disease

reduction of 78% under greenhouse conditions [141]. On the other hand, in greenhouse assays, the commercial product Serenade Max, corresponding to the *Bacillus subtilis* QST713 strain, considerably reduced the incidence and severity of the disease [164]. It should be considered that for the detection of antagonist bacteria with biocontroller potential, it is necessary to perform both laboratory and field assays independently, to determine their effectiveness [132]. A strain with proved biocontroller potential in vitro may not have the same results in field trials; an example of this is the case of the antagonist strain *P. syringae* (22d/93) and its two antibiotic-resistant derived mutants, which in field trials, despite expectations, did not significantly control *P. syringae* pv. *glycinea* infection in soybean plants [165]. Currently, the BCAs applied in agriculture to manage *P. syringae* correspond mainly to bacteria and fungi. However, recently, bacteriophage viruses have acquired renewed interest as a promising alternative that seeks to alleviate the shortcomings of other strategies. In the next section, the biocontrolling potential of these viruses with natural bactericidal activity against *P. syringae* will be summarized.

5. Bacteriophages in *Pseudomonas syringae* Control

One of the most promising strategies for the management of plant diseases caused by bacteria is the use of specific bacteriophages [76]. Bacteriophages or phages are viruses that infect bacteria and are the most abundant biological entity in the biosphere [166], with a vast genetic and morphological diversity. According to the International Committee on Taxonomy of Viruses (ICTV), there are 165 phage species that infect members of the genus *Pseudomonas* (*Pseudomonas* virus). However, this number could be widely exceeded if it is considered that not all the phages examined are sequenced and deposited in public databases, and therefore it is possible that several phages remain unsorted. Of the species listed by the ICTV, a vast majority belong to the order *Caudovirales*, 7 to the *Cystoviridae* family, 2 to the *Inoviridae* family and 2 to the *Tectiviridae* family (https://talk.ictvonline.org/taxonomy/, accessed on 9 August 2021). The genome of *Pseudomonas* viruses is made up of DNA or RNA, which in turn can be double or single-stranded and is packaged in a capsid that can be polyhedral (*Tectiviridae*, and *Cystoviridae*), filamentous (*Inoviridae*), or connected to a tail (*Caudovirales*). Within the *Caudovirales* order, *Pseudomonas* viruses are classified into the families *Autographiviridae*, *Myoviridae*, *Podoviridae,* and *Siphoviridae*. Morphologically, the virions of the families *Autographiviridae* and *Podoviridae* are characterized by a short non-contractile tail; *Myoviridae* is characterized by the presence of a rigid and contractile tail, and *Siphoviridae* by a long and flexible tail (https://talk.ictvonline.org/taxonomy/p/taxonomy_releases, accessed on 9 August 2021). Depending on the replicative cycle, bacteriophages can be subdivided into lytic and lysogenic bacteriophages. Phages using the lytic pathway are considered virulent, and by definition, infection with a lytic phage will result in the lysis of the host, followed by the release of phage progeny. On the other hand, temperate-type phages can follow both the lytic and lysogenic route to infection. In the latter, the phage genome integrates into the bacterial chromosome or persists as a plasmid. In this way, the phage is known as a prophage, and thus replicates as part of the bacterial genome until a triggering factor induces a change in the lytic cycle [167].

The use of phages as agents with antimicrobial potential began in the 1920s, early after their discovery, independently, by F. Twort in 1915 and F. D'Herelle in 1917. Although the first applications of phages were focused on humans [168] (bacteriophage therapy), other fields, including agriculture, soon began to explore their potential as biological control agents (i.e., bacteriophage biocontrol). The first experimental evidence that phages could be associated with plant pathogenic bacteria was presented in 1924, when it was shown that a filtrate obtained from decaying cabbage could inhibit the cabbage rot caused by *Xanthomonas campestris* pv. *campestris* [169]. The following year, Kotila and Coons [143] demonstrated that exposure of *Pectobacterium atrosepticum* and *P. carotovorum* subsp. *carotovorum* to phages could prevent soft rot in potato and carrot tuber slices, respectively [170,171]. The first recorded field test was in 1935, when Stewart's disease of corn, caused by *Pantoea stewartii*, was reduced by phage pretreatment of seeds, thus demonstrating its

effectiveness [172]. However, these important advances in the application of bacteriophages were unconvincing in terms of efficacy and reliability; furthermore, their biological nature was poorly understood [173]. Phage therapy trials in the United States and most of western Europe ceased after World War II, and research in this area was displaced by the discovery of broad-spectrum antibiotics in the 1940s [168]. At that time, the widespread application of antibiotics made it possible to manage phytopathogenic bacteria and treat diseases both in humans and in animal husbandry [15].

Currently, the incorporation of bacteriophages as biocontrol agents has regained interest, gaining relevance in human and veterinary medicine [161,174], aquaculture [175] and food safety [176], with studies that demonstrate its efficacy even in antibiotic-resistant bacteria [177]. Likewise, in recent years, numerous reviews on biocontrol with bacteriophages in agriculture have been published [2,16,18,76,79,80,178], in which promising results are compiled on a series of important diseases caused by phytopathogenic bacteria in potato, tomato, grape, onion, lettuce, radish, grapefruit, orange, leek, and mushroom plants, and apple trees, among others. Table 3 summarizes the most relevant milestones that have driven and renewed interest in the development of phage therapy for use in agriculture.

Table 3. Timeline of the development of phage therapy against *Pseudomonas syringae*.

Year	Milestone	Reference
1915–1917	Phage discovery	[179]
1924	First isolation of phages that infect phytopathogenic bacteria	[169]
1935	First field test demonstrating the effectiveness of phages to treat seeds infected with Stewart's wilt	[172]
1943	Discovery of streptomycin and the beginning of the golden age of discovery and development of antibiotics (1940–1990)	[180]
1979	First report of streptomycin resistance in a phytopathogenic bacterium [1]	[181]
1984	First report of resistance to copper in a phytopathogenic bacteria [2]	[182]
2004	Banning of the agricultural use of streptomycin in the European Union	[183]
2005	The first commercial pesticide containing bacteriophage (AgriPhage ™) against *Pseudomonas syringae* pv. *tomato* and *Xanthomonas campestris* pv. *vesicatoria* is registered in the USEPA	[135]
2014	Isolation and characterization of the first specific bacteriophages against *P. syringae* pv. *actinidiae* with potential application in biocontrol	[24,184]
2016	Isolation, characterization, and evaluation in field tests of the first specific bacteriophages against *P. syringae* pv. *porri*	[25]
2020	Publication of the first study demonstrating the ability of a phage cocktail to reduce the *P. syringae* pv. *actinidiae* load on kiwi trees in vivo.	[27]
2020	Isolation, characterization, and in vivo evaluation of the first specific bacteriophages against *P. syringae* pv. *syringae and* pv. *morsprunorum*	[134]

[1] *Erwinia amylovora* strains isolated in California, USA, harboring a chromosomal resistance mutation to streptomycin. [2] Resistance conferred by a plasmid identified in *Xanthomonas campestris* pv. *vesicatoria*.

The advantage of phages that distinguishes them from other BCAs is their capacity for self-replication and self-limitation, since they replicate only while the host bacteria are present in the environment, and their population decreases rapidly in its absence [76]. In addition, they can be isolated from anywhere bacteria are present, including soil, natural and sewage water, plants, animals, and even the human body [185]. Due to their narrow hosts (high specificity)—which can range from the ability to infect only a few strains of a bacterial species to, very rarely, the ability to infect bacteria belonging to different genera considered relatively close—phages can be considered harmless to beneficial members of the native microbiota [186]. Phages are not toxic to the eukaryotic cell, so they can be implemented in situations in which chemical management is restricted. It has been seen that phages can mix with other agrochemicals without a significant loss of viral titer [187,188] and in addition, they are capable of degrading bacterial biofilms [189–191]. Even if phage-resistant bacteria emerge, they may become less virulent, as this resistance may be due to the loss of function of the bacterial receptors essential for pathogenesis [76]. Despite the advantageous properties of bacteriophages and numerous investigations on

this subject, only a small number of studies have resulted in the development of commercial phage-based products for agricultural use. In the case of *P. syrinage*, it should be mentioned that phage studies of this bacterium date back at least to the 1970s [192,193]; however, not all of them focused on demonstrating the usefulness of phages as BCAs. Some of these studies looked for transducing phages (temperate phages), and others used phages for typing bacteria [194,195]. Even so, many studies focused on the use of phages as BCAs only go as far as the isolation and general characterization of these viruses, with only a few studies carrying out field trials and demonstrating the biocontrolling potential of phages in vivo and under the challenging environmental conditions; this constitutes the main limitation of the use of these viruses in agriculture.

According to what is mentioned throughout this review and what is summarized in Table 4, regarding the biocontrol of *P. syringae* with bacteriophages, the most studied pathovars are *actinidiae, morsprunorum, porri, syringae* and *tomato.* Tables 4 and S1 summarize the main results published in the last 50 years, focusing on the isolation and use of phages against *P. syringae*. Although the results are encouraging, the state of progress of the different studies confirms the lack of trials carried out in field conditions. Only in recent years have diverse groups conducted in vivo studies and field assays that reveal the true potential of bacteriophages for use as BCAs against *P. syringae.* First, in 2005, after greenhouse and field tests, a pioneering product was registered with the United States Environmental Protection Agency (USEPA). Developed by Omnilytics, Agriphage™ is the first bacteriophage-containing biopesticide for use in agriculture and is indicated for the control of bacterial spot and bacterial speck on tomatoes and peppers, caused by *X. campestris* pv. *vesicatoria* and *P. syringae* pv. *tomato*, respectively (https://www.agriphage.com/, accessed on 12 February 2023). In this context, almost a decade later, the first publications on the isolation and characterization of new phages appeared, in which their therapeutic potential to control the other pathovars of *P. syringae* also began to be evaluated. In 2014, two independent groups reported the first specific bacteriophages against Psa, the causal agent of kiwi bacterial canker [24,184]. Subsequent studies demonstrated the potential of phages to control Psa in vitro, ex vivo, and in combination with an essential oil [191,196–199]. However, it was not until this year that the first in vivo study was published, demonstrating the ability of a phage cocktail to reduce the Psa load on kiwi leaves by more than 75% [27]. Regarding *P. syringae* pv. *porri*, Rombouts et al. (2016) led an unprecedented study, in which they isolated and evaluated the potential of specific phages against the causal agent of bacterial blight of leek in vitro, in vivo, and in field trials [25]. While in vitro and in vivo tests showed significant and positive results, only one experiment in the field trials showed that a phage cocktail could reduce the incidence of the disease. Finally, a recent study [111] reported the isolation, characterization, and the first in vivo evaluation of phages against *P. syringae* pv. *morsprunorum* and pv. *syringae*, which cause bacterial canker in cherry. The phages proved to be effective in both in vitro and in vivo tests, showing that phages (individually and in cocktails) were able to significantly reduce the bacterial population [134].

Table 4. Summary of published studies which have focused on the use of bacteriophages as biocontrol agents against *Pseudomonas syringae*, during the last 50 years.

Target Pathogen/Host	Assay Type	Principal Results	Reference
Psa/kiwi	Effect of the phage is evaluated by observing necrotic areas in kiwi plants with symptoms of Psa under greenhouse conditions (in vivo assay). Detailed phage characterization was carried out previously.	φPSA2 is effective in preventing Psa replication inside plants, and capable of reducing the number and size of lesions produced by the bacteria. The phage is also capable of killing *Pseudomonas* when present on the leaf surface.	[200]
Pss/lemon	Phages are isolated from soil samples, irrigation water and symptomatic lemons infected with *P. syringae* pv. *syringae*. Bioassays in lemons measure the percentage of necrotic tissue (in vivo assay). HR (12), TEM, ST, OSGC (MOI:0.01) and GS.	In the bioassays, SoKa reduced the symptoms of infection, but could not prevent it.	[201]

Table 4. *Cont.*

Target Pathogen/Host	Assay Type	Principal Results	Reference
Psa/kiwi	Hairong and ZY21 are isolated from Psa-infected symptomatic plant tissue by the soft agar plaque method (in vitro assay). HR (31), TEM and GS.	Hairong and ZY21 have relative phylogenetic closeness to two nickie-like phages (psageB1 and nickie) based on major capsid protein sequences.	[202]
Pst/pepper	Bacteriophages are isolated from peppers that exhibit symptoms of Pst infection. In *A. thaliana*, Pst or the mixture of the bacteria with the phages is inoculated (in vivo assay). HR (8), TEM, AC, KCA (MOI:0.01), ST and GS.	In vivo, the co-inoculation of Eir4 and Eir9 requires a low MOI to obtain effective phage propagation and Pst inactivation. Even so, the separate treatment, in comparison to the plants with only Pst, resulted in leaves yellowing less, and showing an almost normal growth.	[203]
Psa/kiwi	The phages were isolated in a kiwi orchard from canker branches or soil suspension. The lytic activity of the phage cocktail was determined, and individual phages were Psa inoculated in red-fleshed kiwifruit seedlings (in vivo assay). TM, OCGC, ST, KCA (MOI:1) and RFLP.	The phage cocktail in the infected plant generated an increase in phage viral particles during the first 12 h; however, the determined phages had a significant increase at 72 h, thus verifying the superior effect of the phage cocktail.	[204]
Pss/green bean	Pf-10 phage is isolated from tissue infected with Pss of green bean. HR (7), TEM, ST, AC, KC (MOI:0.1), OSGC, RLFP and GS.	Pf-10 genome is a linear dsDNA that contains 49 genes. Presents a variety of endolysins and putative holins.	[205]
Psa/kiwi	Phage was isolated from soil samples of "hongyang" kiwi crops. The efficacy of PHB09 is evaluated on leaf discs of kiwi plants (in vivo assays). HR (6), TEM, OSGC (MOI:0.001), ST and GS.	In kiwi leaves with Psa, a decrease in the bacterial load is observed and the symptoms do not occur.	[206]
Psa/kiwi and Pph/bean	The isolated phages were obtained from plant, soil and wastewater samples close to plants infected with Psa and Pph that presented symptoms. HR (32), TEM, ST and GS.	The phages exhibited selective killing of pathogenic *Pseudomonas* strains in in vitro assays; however, psageB1 lysed three non-pathogenic strains.	[207]
Pss/cherry	Isolation and in vitro determination of lytic activity using the spot inoculation method against *P. syringae* pathovars. The effects of bacteriophages against Pss were determined in micro propagated cherry plantlets in vivo and under growth chamber conditions.	Results of in vivo assays performed in cherry plantlets demonstrated that at 10 days post inoculation, 4 out of 6 phage treatments (F1226, F137, F358, F369) successfully reduced more than 50% of the disease incidence caused by the high-virulence *Pss* strain BY5L316.	[208]
Psa/kiwi	Isolation and in vitro determination of lytic activity using the spot inoculation method. HR (29), TEM, OSGC (MOI:0.01), ST and GS.	PN09 showed lytic activity against the 29 Psa biovar 3 strains tested. PN09 showed specificity for Psa and did not lyse other bacterial species tested.	[191]
Psa/kiwi	Control efficacy of PPPL-1 phage alone and in combination with KHUφ34 and KHUφ38 against bacterial canker was tested in vivo in kiwifruit plants under greenhouse conditions.	Results showed that the disease control efficacy of PPPL-1 treatment was statistically similar to that of the phage cocktail (mix of three phages) treatment or an agrochemical containing streptomycin and oxytetracycline antibiotics as active ingredients.	[198]
Pss/unspecified host	The bacteriophage was isolated from irrigation water on a farm where tomatoes were grown. In vitro determination of lytic activity using the soft agar plaque method. HR (17), TEM and GS.	Host range analysis showed that 64.7% of the bacterial strains investigated were susceptible to the phage Phobos, including *P. syringae* pathovars *syringae* and *tomato*. Sequence analysis of the predicted proteins encoded by the Phobos genome showed no homology to known virulence factors, antibiotic resistance factors, or potential immunoreactive allergens.	[26]
Pspo/leek	The overall performance of a cocktail containing both phages was assessed in a seed bioassay at MOI:10. BR. Detailed phage characterization was carried out previously.	A combination of KIL3b and KIL5 phages reduced the bacterial concentration 100-fold in seed bioassay. In vitro Pspo resistance against phage infection developed quite rapidly; however, the virulence of those mutants is possibly reduced.	[209]
Pae/horse chestnut tree	For phage isolation, soil and leaf samples of healthy and diseased trees were used, and in vitro determination of lytic activity was carried out using the soft agar plaque method. Co-evolution experiments were also performed. HR (22), TEM, RAPD, BR and KCA (MOI:0,1).	Most phages were able to infect all the tested *P. syringae* pv. *aesculi* (2250, 6617, 6619, 6620, 6623, 6631), alongside another *Pseudomonas* (*P. syringae* pv. *lachrymans*, *P. syringae* pv. *tomato*, *P. marginalis* and pv. *marginalis*). In the best case, a reduction of approximately 65% in the bacterial growth was observed at 24 h in the KCA.	[22]
Ps/unspecified host	Phages targeting *P. syringae* GAW0113 were isolated from organic waste samples. HR (13), EOP, TEM and GS.	All three phages were found to infect different strains of *P. syringae* covering several phylogroups. Three phages were shown to have a narrow host range, infecting 3 out of 13 *P. syringae* strains.	[210]
Psa/kiwi	Phages (PN05 and PN09) were isolated from water samples. A phage combined with varying concentrations of carvacrol was added to a Psa inoculum at an MOI: 1 for the different in vitro experimental setups. KCA (MOI: 0,1, 1, 10 and 100) was performed to characterize phages.	The combined treatment of phages and carvacrol (2.0 mg/mL) showed a higher efficacy (in relation to phage therapy or carvacrol alone), reducing (by 5.87 log CFU/mL) and preventing Psa regrowth for more than 40 h.	[191]

Table 4. *Cont.*

Target Pathogen/Host	Assay Type	Principal Results	Reference
Psa/kiwi	Phage φ6 (DSM 21518) was tested against two biovar 3 strains (Psa CRA-FRU 12.54 and Psa CRA-FRU 14.10). The inactivation of Psa was assessed in vitro using liquid culture medium, and ex vivo using artificially contaminated kiwifruit leaves. AC, OSGC and KCA (MOI:1)	In the in vitro experiments, phage φ6 was effective against both tested strains (maximum reduction of 2.2 and 1.9 CFU/mL for Psa CRA-FRU 12.54 and Psa CRA-FRU 14.10, respectively). In the ex vivo tests, the decrease was lower (maximum reduction 1.1 log and 1.8 CFU/mL for Psa CRA-FRU 12.54 and Psa CRA-FRU 14.10, respectively).	[199]
Pss and Psm/cherry trees	Phages were isolated from the soil, leaf, and bark of cherry trees. In vitro determination of lytic activity was carried out the soft agar plaque method. In vivo assays were performed in bean plants and cherry trees using leaf or twig inoculation with Pss and Psm. In both cases, a MOI:0.01 was used. HR (22), KCA, TEM, RAPD-PCR, GS, ST and BR.	In bean leaves, the best results were obtained with individual phages MR6 and MR7, which reduced the bacterial population (Pss) by 50%. The bioassays performed in cherry leaves showed that phage MR16 reduced the bacterial population to almost zero, and phage cocktails reduced the Pss bacterial population by 50%. In cherry twig inoculation assays, all phages, both individually and in phage cocktails, reduced the bacterial population. The best results were obtained in the case of phage MR8, which reduced the growth of all three bacteria by 60%.	[134]
Psa/kiwi	The phages were isolated from soil and water samples using different strains of Psa biovar 3 obtained from Chilean kiwifruit orchards as the host. Ex vivo assays were performed using kiwifruit leaf discs. Moreover, in vivo experiments were performed with two-year old kiwifruit plants cultivated in greenhouse conditions. A MOI:10 was used in the different performed bioassays. HR (18), KCA, TEM, RFLP, ST, GS and BR.	Under laboratory conditions, with kiwifruit leaf samples, the results showed that a cocktail of phages CHF1, CHF7, CHF19, and CHF21 reduced the bacterial load below the detection limit (20 UFC/mL), even 24 h post inoculation. In addition, the treatment with the phage cocktail was able to protect kiwifruit leaf discs from the damage produced by Psa. In the in vivo experiments, the phage cocktail was able to reduce the Psa load by more than 75%, in comparison with the untreated plants. Moreover, the damage index decreased from 2.3 (without phage treatment) to 1.3 (treated with phage cocktail).	[27]
Pss/unspecified host	In vitro characterization of bacteriophage φ6 (DSM 21518) lytic activity against bacterial strains of *P. syringae* pv. *syringae* and other bacterial strains of interest. HR (25), ST, OSGC.AC, KCA (MOI:1 and MOI:100) and BR.	The host range analysis revealed that the phage, besides its host (*P. syringae* pv. *syringae*), also infects the *P. syringae* pv. *actinidiae* CRA-FRU 12.54 and CRA-FRU 14.10 strains, not infecting strains from the other tested species. An MOI 1 (maximum reduction of 3.9 log CFU/mL) was more effective than MOI 100 (maximum reduction of 2.6 log CFU/mL) in deactivating the bacterium.	[78]
Psa/kiwi	PPPL-1 was isolated from soil of a kiwifruit orchard. The lytic activity of PPPL-1 was determined in vitro against *P. syringae* pv. *actinidiae* strains and strains from other pathovars, including *aptata, syringae, tomato, glycinea, phaseolicola, pisi* and *tabaci*, among others. HR (53), KCA (MOI:0.01), ST and GS.	PPPL-1 showed specificity for *P. syringae* species and was effective against 16 of the 18 tested Psa strains. PPPL-1 can maintain its lytic activity against Psa strain KBE9 stably for at least 80 h.	[198]
Pspo/leek	Phages were isolated from soil samples from the same fields from which the *P. syringae* pv. *porri* strains were taken. In vitro assessment of the phages' lytic activity against Pspo strains was carried out according to the soft agar overlay plate technique. In vivo bioassays and field trials were performed. The activity of phages was tested in vivo (MOI: 100) by injecting phage and bacterial suspensions into leek leaves. HR (46), TEM, KCA, AC, ST, BR and GS.	None of the phages infected all the *P. syringae* pv. *porri* strains tested, but the combined host range of the phages covered all 41 Pspo isolates tested. In vivo bioassays showed that the phages KIL1, KIL2, KIL3, and KIL3b are able to reproduce inside the plant tissue, and lead to a significant reduction in the lesion length when coinjected with the bacterial host. However, the effect of phages KIL1, KIL2, and KIL3 varied between the assays.	[25]
Psa/kiwi	Bacteriophages against *P. syringae* pv. *actinidiae* were isolated from soils collected from kiwifruit orchards. HR (31), TEM, KCA (MOI: 0.01), DGREA and ST.	Bacteriophage KHUφ44 was the only phage effective against all 18 Psa strains tested, but it had only limited effects on two of them. The combined host range of the phages covers all 18 Psa strains tested. Most of the bacteriophages were also effective against other *P. syringae* pathovars (*tabaci, tomato* and *phaseolicola*), and none showed effect on other bacteria. The lytic activity of bacteriophages KHUφ34, KHUφ38 and KHUφ44 was sustained in vitro until 80 h.	[197]
Different bacteria genera and species, including *P. syringae* spp.	Isolation from sewages samples. The main objective was finding polyvalent phages and a method to obtain those phages. TEM, HR (7), AC, OSGC and KCA (MOI:10).	Phages with multiples host tropism were obtained. Lytic phages were capable of interspecies or inter-order infectivity without a significant reduction in plating efficiency. Phage PX1 delayed the onset of exponential growth for each host by 3 h and reduced the maximum viable bacterial density (CFU reaching stationary phase) by 50% for *P. syringae*.	[211]
Psa/kiwi	Bacteriophages were obtained from leaves of *A. deliciosa* infected by Psa, and in vitro determination of lytic activity was carried out using the soft agar plaque method. TODHR (51), TEM, LF, AC, OSGC (MOI:0.01), ST and GS.	φPSA2 is a strictly lytic phage and exhibits a broad host range, being lytic against all the 37 Psa strains tested and some other pathovars including *theae, avellanae* and *morsprunorum*.	[184]

Table 4. *Cont.*

Target Pathogen/Host	Assay Type	Principal Results	Reference
Psa/kiwi	Samples for phage isolation consist of soil, water, and leaf litter collected from infected kiwifruit orchards. Lytic activity was determined using the soft agar plaque method. HR (32), TEM, ST, BR, LF, T, DGREA and GS.	The host range of individual phages was narrow, but all the Psa strains tested were infected by at least one of the isolated phages. In total, approximately 20,000 phage–host combinations were examined, and showed clear differences in the phage profiles of *P. syringae* pv. *Actinidiae* strains from distinct geographic locations.	[24]
Pst/tomato	16 phages were isolated from tomato field soils and plant debris from various locations throughout Ontario. HR (106), LF, RTD, TEM and ST.	Over 70% of the Pst strains were lysed by a group of 13 PT phages in in vitro assays. Phages PT1, PT18, PT20 and PT32 showed a high degree of specificity for Pst virulent strains, and were able to infect 89, 89, 82 and 87% of the tested Pst strains, respectively, including strains from Australia, New Zealand, Europe, and the USA.	[23]
Psg/soybean	Phages were isolated from raw sewage obtained from four resources in Riverside and San Bernardino counties in California. Phages' specificity for different *P. syringae* pathovars was determined in vitro. HR (32), OSGC, TEM and T.	The phages isolated were virulent on most of the pathovar *glycinea* strains. Altogether, 6 of the 7 selected phages were able to infect most of the pathovars tested, including *lachrymans*, *morsprunorum*, *phaseolicola*, *pisi*, *savastanoi*, *tabaci* and *tomato*. Only the phage R4-0B was specific for the pathovar *glycinea*. The 7 phages proved to be specific for *P. syringae*, being capable of infecting 30 of the 33 tested strains belonging to this species.	[212]
Psm/cherry trees	Isolation and in vitro characterization of a phage specific to Psm race 2. The phage's lytic activity was assessed via RTD. The phage was used in a survey of *P. morsprunorum* races isolated from commercial orchards and from the cherry cultivars Napoleon and Roundel in a research station. HR (134).	The data showed that 55 of the 134 tested *Pseudomonas* strains were susceptible to B1 phage, all of them belonging to the *morsprunorum* pathovar, and 52 belonging to race 2.	[193]

Abbreviations used to describe bacteriophage characterization are as follows: HR (n): host range (number of total tested bacterial strains). Methods of HR determination could differ between different studies. LF: lysogenization frequency; AC: adsorption curves; OSGC: one step growth curves; ST: stability tests; T: transduction assay; BR: phage-resistant bacteria frequency; GS: genome sequencing; DGREA: direct genome restriction enzyme analysis; RTD: routine test dilution; TEM: transmission electron microscopy; RAPD: random amplification of polymorphic DNA by PCR; KCA: killing curves assay, TOD: time of dead (time required for reduction of the culture optical density from 0.2 to 0.1.) and RFLP: restriction fragment length polymorphism analysis.

Despite the advantageous characteristics of phages, there are challenges that should be considered for their use as BCAs in agriculture. Among them, the sensitivity of phages to environmental factors stands out, affecting their persistence in the plant environment. The phyllosphere environment is harmful to phages, leading to population decline over time. This low persistence on plant leaf surfaces is the main limiting factor to phage therapy in the phyllosphere; it is influenced by desiccation, temperature, pH, and above all, sunlight irradiation (especially in the UV spectra A and B) [16]. On the other hand, in rhizosphere environments, the low diffusion of phages through the diverse soil matrix, their pH, and humidity can limit their activity as BCAs [179]. Another relevant aspect is the problematic use of temperate phages due to the possibility of alternating a lytic cycle with a lysogenic cycle. In such a case, the risk is higher, considering the existence of immunity to superinfection, which renders phage-sensitive bacteria insensitive, and the possibility of temperate phages encoding for bacterial virulence factors, making them capable of converting harmless bacteria into pathogens [186]. Constant exposure to phages exerts a selective pressure that favors the emergence of bacteria resistant to their infection [213], with cross-resistance being the most undesirable scenario. Furthermore, although the host range of a phage is generally narrow, there is still the possibility that it could infect a native or beneficial strain. Long-term storage can compromise the infective capacity of virions [214]. Table 5 and Figure 3 summarizes the principal considerations that should be taken when implementing a bacteriophage biocontrol program and the possible strategies to overcome these limitations.

Another issue to consider is the possibility of integration of phage-based treatments in 'smart farming' programs, allowing early detection of infection and targeted phage application to contain the disease. Smart farming takes advantage of technologies based on artificial intelligence (AI) (smartphones, robotics, machine learning and sensor-based technologies) to monitor crops, which have proved to be helpful in the detection of different

phytopathogens, both in greenhouse and field conditions. In this context, the rational application of resources is both economic and ecological logical, as it reduces the number of treatments needed. For example, AI-powered sensors and drones could be useful to monitor crop health and detect diseased crops in open field situations, even at an early stage. In this sense, AI can help farmers to take action before the problem becomes too severe, since diseased crops can be detected and eradicated in the field in an efficient manner, while neighboring plants are treated with a biopesticide [220].

Table 5. Main limitations for phage-based management application in agriculture and possible strategies to overcome them.

Limitation	Strategies
Phage persistence in the phyllosphere and rhizosphere (formulation and mode of application)	In response to this problem, protective formulations have been investigated to minimize UV damage, although there is a great need to identify effective formulations. Based on self-replication ability phage survival can be improved in the phyllosphere and rhizosphere if they are accompanied by a viable host [2]. As an alternative, it is worth considering artificial phage evolution to increase resistance to UV-induced damage [215] Phage delivery through the soil is another approach that has been explored to improve phage persistence in the phyllosphere. There is a phage translocation pathway from the roots to the leaves of plants through the vascular system of the plant, possibly via xylem. Specifically, it has been shown that phages can translocate in tomato, rice, apple, and fire thorn plants [216–218]. It is suggested that if the phages can translocate systemically in the plant, then they could possibly be used therapeutically after infection by a bacterial pathogen by applying the phages to the surrounding soil of a plant instead of foliar spray [2].
Potential alteration in the phage replication cycle (lytic to lysogenic)	To avoid this problem, ideally a phage for biocontrol applications should be exclusively lytic. Preferably, phages that produce transparent plaques should be chosen to reduce the isolation of temperate phages since the latter can carry out the unwanted lysogenic conversion [2,79]. Lysogens contain a prophage and are typically resistant to reinfection by the same phage, which results in turbid plaques via superinfection immunity [24]. Currently, there are validated protocols to assess whether a phage is lysogenic or if it is capable of transferring genes between bacteria (transduction test) [24], allowing to rule out those that present a risk for their use as BCAs. Additionally, it is necessary to analyze the genome of the phages to be used, discarding those with genes encoding for bacterial virulence factors or antimicrobial resistance genes, among others [27].
Phage resistant bacteria	To avoid the problem of the high frequency of bacteria resistant to phage treatment, a combination of phages with different infection mechanisms can be used, reducing the probability of the appearance of resistance [80]. Increased diversity within the known phages targeting *P. syringae* strains also allow for development of more complex phage cocktails [210]. It should be considered that even when resistance develops, it can lead to a great cost of fitness that entails a deterioration in virulence or a reduction in the growth rate, thus reducing the severity of the disease [80]. Furthermore, in 1989, a patented process was developed to prevent the emergence of phage-resistant mutants [219].
Low efficacy and consistency of control compared to conventional treatments.	Studies have shown that the timing of bacteriophage application is essential to extend the persistence of high populations in the vicinity of the host bacteria to promote biological control [18]. A strategy for a phytosanitary program should include phage applications throughout the season to reduce the population of pre-existing pathogens or avoid its proliferation [16]. Another key factor for the effectiveness of the use of phages is the time of day they are applied since ultraviolet light from the sun is capable of inactivating viruses. A possible strategy consists of night applications on the leaves, managing to increase the persistence of the phages in the phyllosphere and thus their bactericidal action [2]. The use of formulations with phages could complement other strategies for disease management, as part of an integrated phytosanitary program for pest management, increasing effectiveness and sustainability. These strategies would include hypersensitivity and systemic resistance response activators, copper-based agrochemicals, or antibiotics such as streptomycin. In this sense, it would be possible to minimize the probability of selection for resistance to these components or to phages [24].

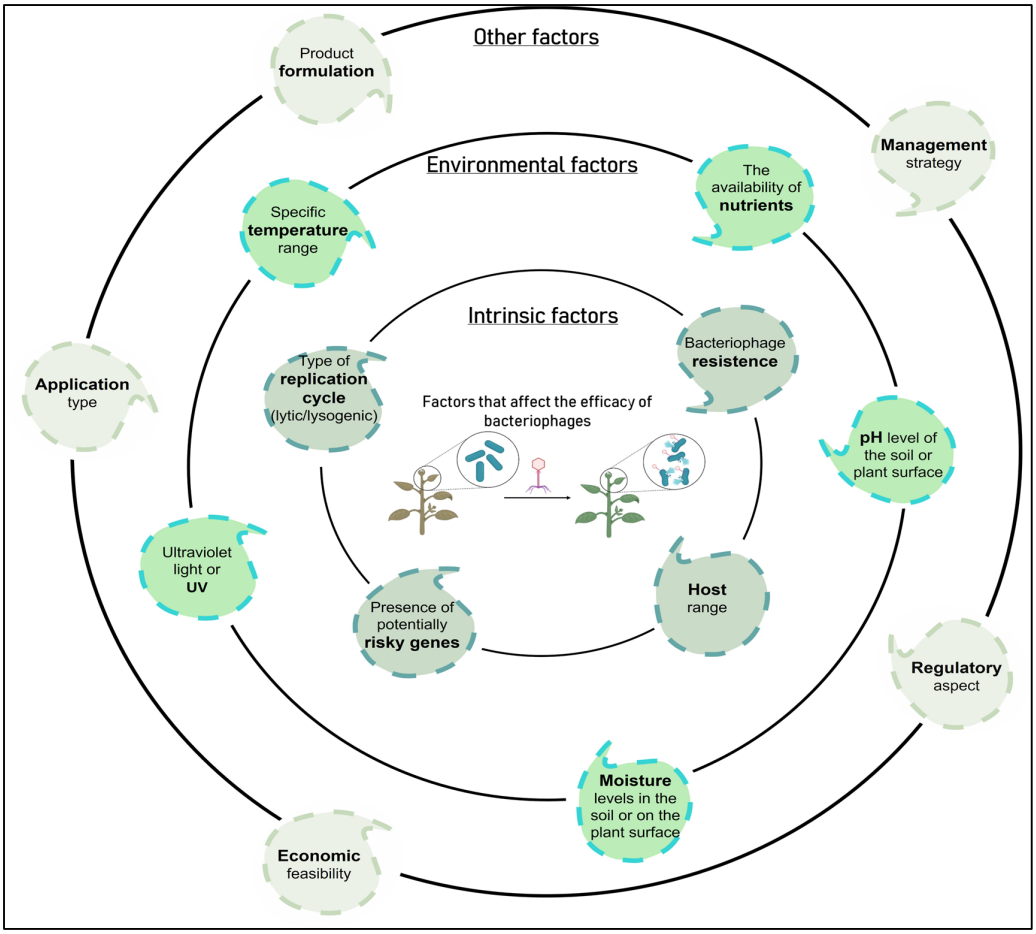

Figure 3. Levels of factors to consider for the development of a control of phytopathogenic bacteria with bacteriophages.

In summary, while biological control offers sustainable and environmentally friendly solutions for pest and disease management in agriculture, it is not without its challenges and trade-offs. Overcoming these challenges and making informed decisions regarding the use of biological control agents requires a comprehensive understanding of the specific agricultural system in question, the target pests or diseases, and the available biological management options. By addressing these challenges and optimizing biological control strategies, it is possible to enhance the effectiveness and adoption of biocontrol in agriculture. Table 6 discuss some of these challenges and trade-offs.

Table 6. Principal challenges and trade-offs in biological control in agriculture.

Challenges	Description
Efficacy	One of the primary challenges of biological control is ensuring its effectiveness in controlling pests and diseases. The success of biological control agents depends on several factors, including the target pest or disease, the specific biological control agent used, and the environmental conditions. Some biological control agents may not be as effective as chemical pesticides, and their efficacy can vary depending on the circumstances. It is crucial to identify and develop biological control agents that are highly specific to the target pest or disease to maximize their efficacy.
Compatibility	Biological control agents are living organisms, and their interactions with the target pest, the crop, and the environment can be complex. Ensuring compatibility between the biological control agents and the agricultural system is crucial. Factors such as temperature, humidity, and pesticide use can influence the survival and efficacy of biological control agents. It is necessary to carefully assess and optimize the conditions under which biological control agents are deployed to achieve the desired outcomes.

Table 6. *Cont.*

Challenges	Description
Cost-effectiveness	Biological control can sometimes be more expensive than chemical pesticides. Developing and mass-producing biological control agents can involve substantial costs, and their application may require specialized equipment or additional labor. Additionally, biological control often requires a longer period to achieve control compared to chemical pesticides. Farmers need to consider the cost-effectiveness of biological control in relation to their specific crop, pest, and economic conditions.
Regulatory challenges	The use of biological control agents in agriculture is subject to regulations to ensure their safety for humans, non-target organisms, and the environment. These regulations may vary across countries and regions. Obtaining necessary approvals and meeting regulatory requirements can be time-consuming and costly. It is essential to navigate the regulatory landscape and comply with the necessary guidelines to use biocontrol agents legally and responsibly.
Knowledge and expertise	Implementing biological control strategies effectively requires knowledge and expertise. Farmers need to understand the biology, behavior, and application methods of biological control agents. They must also be able to monitor pest populations, assess the impact of biological control, and make informed decisions regarding their use. Providing training and support to farmers to enhance their understanding and skills in biocontrol is crucial for successful implementation.
Trade-offs	There can be trade-offs associated with biological control in agriculture. For example, certain biological control agents may have a narrower range of effectiveness compared to chemical pesticides, meaning they may only target specific diseases. This specificity can be advantageous in reducing non-target effects, but it may require the use of multiple biological control agents for different diseases, thus increasing complexity and costs. Additionally, biological control agents may not provide immediate control, and may require more consistent monitoring and application compared to chemical pesticides.

6. Concluding Remarks

The plant pathogen *P. syringae* constitutes a continuous threat to the agricultural industry, causing great economic losses in recent decades in various countries [46,47]. Current management strategies against *P. syringae* have been less and less effective, and bacterial resistance to agrochemicals is a significant problem [77,96,97,99]. Despite constant efforts to manage this pathogen, there is still no effective and sustainable strategy for its management [68,73,78]. Given its harmful effects on human health, the environment, and even on agricultural crops with conventional chemical treatments, the last few years have seen focused efforts to develop ecological alternatives for the management of this pathogen [2,220]. In the field of biocontrol, different options have been investigated, including the use of fungi, bacteria, and bacteriophages, with encouraging but variable results. To elucidate the conditions conducive to the success or improvement of biological control strategies, it is necessary to understand the mechanisms of the interactions between biocontrol agents and pathogens; among them, we can find direct mechanisms such as competition, secondary metabolite production (for example, antibiotics), or bactericidal properties (bacteriophages) [90,159,220], and indirect mechanisms such as ISR [94,132,145,160]. Among potential biocontrol agents, bacteriophages appear to be one of the most promising alternatives, due to their various advantages. However, studies have focused mainly on their isolation and characterization, with only a few trials being carried out in field conditions (Table 4). Of particular interest is the use of phages in the nursery for improving the production and commercialization of woody plants and seeds free of phytopathogenic bacteria. In this sense, it is essential to focus efforts on consolidating knowledge on management strategies based on bacteriophages which are suitable for application in the field and in different environmental conditions. This need has been reflected in more recent studies focused on evaluating the stability of different bacteriophage formulations under different conditions that simulate the conditions in which they will be applied [221,222], these studies have concluded that it is possible to design a strategy

that is compatible with the use of phage therapy in agricultural environments [222,223]. However, it must be considered that different types of orchards or crops will offer different environmental conditions or challenges within phage application. Therefore, the process of optimization of a phage-based product may vary depending on the fruit or vegetable produced. In addition, it must be considered that there are some commercial products based on bacteriophages that have already been successful [220], so the prognosis for this technology is promising.

In pursuit of an effective and environmentally friendly form of integrated pest management, it is reasonable to think about the use of biocontrol agents in combination with other biocontrollers or chemical treatments, exploiting their different attributes and advantages for the prevention and management of diseases caused by both *P. syringae* and other plant pathogens [95,126,191]. However, the compatibility of phages with other BCAs or agrochemicals has not been thoroughly examined, and tests of viability and persistence would be required in the presence of any treatment used simultaneously on affected plants. The potential of bacteriophages as biocontrollers is reflected in the scientific publications that appear every year, showing their effectiveness in the management of different bacterial diseases [2,223]. This new knowledge will allow us to advance in the development of new strategies and will allow us to overcome the initial limitations involved in the use of phages as biological agents in agriculture conditions [18,79,179,222]. Moreover, the integration of advanced technology in the implementation of phage-based management strategies could play an important role in the success of disease prevention and control. In this sense, the use of AI in agriculture has the potential to revolutionize the application of integrated management strategies in agriculture by enabling farmers to make better decisions, thereby improving crop yields and increasing efficiency in farming practices.

Supplementary Materials: The following supporting information can be downloaded at: https://www.mdpi.com/article/10.3390/horticulturae9060712/s1, Table S1: Summary of basic characteristics of phages from published studies focused on the use of bacteriophages as biocontrol agents against *Pseudomonas syringae* over the last 50 years.

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References

1. Kennelly, M.M.; Cazorla, F.M.; De Vicente, A.; Ramos, C.; Sundin, G.W. *Pseudomonas syringae* Diseases of Fruit Trees: Progress toward Understanding and Control. *Plant Dis.* **2007**, *91*, 4–17. [CrossRef]
2. Buttimer, C.; McAuliffe, O.; Ross, R.P.; Hill, C.; O'Mahony, J.; Coffey, A. Bacteriophages and Bacterial Plant Diseases. *Front. Microbiol.* **2017**, *8*, 34. [CrossRef] [PubMed]
3. Hirano, S.S.; Upper, C.D. Bacteria in the Leaf Ecosystem with Emphasis on *Pseudomonas syringae*—A Pathogen, Ice Nucleus, and Epiphyte. *Microbiol. Mol. Biol. Rev.* **2000**, *64*, 624–653. [CrossRef] [PubMed]
4. Monteil, C.L.; Yahara, K.; Studholme, D.J.; Mageiros, L.; Méric, G.; Swingle, B.; Morris, C.E.; Vinatzer, B.A.; Sheppard, S.K. Population-Genomic Insights into Emergence, Crop Adaptation and Dissemination of *Pseudomonas syringae* Pathogens. *Microb. Genom.* **2016**, *2*, e000089. [CrossRef]

5. Monteil, C.L.; Cai, R.; Liu, H.; Mechan Llontop, M.E.; Leman, S.; Studholme, D.J.; Morris, C.E.; Vinatzer, B.A. Nonagricultural Reservoirs Contribute to Emergence and Evolution of *Pseudomonas syringae* Crop Pathogens. *New Phytol.* **2013**, *199*, 800–811. [CrossRef]
6. Ruinelli, M.; Blom, J.; Smits, T.H.M.; Pothier, J.F. Comparative Genomics and Pathogenicity Potential of Members of the *Pseudomonas syringae* Species Complex on *Prunus* spp. *BMC Genom.* **2019**, *20*, 172. [CrossRef]
7. Vanneste, J.L.; Yu, J.; Cornish, D.A.; Oldham, J.M.; Spinelli, F.; Pattemore, D.E.; Moffat, B.; D'Accolti, A. Survival *of Pseudomonas syringae* pv. *actinidiae* in the Environment. *Acta Hortic.* **2015**, *1095*, 105–110. [CrossRef]
8. Pscheidt, J.W.; Moore, L.W. Diseases Caused by Pseudomonas syringae. Available online: https://pnwhandbooks.org/plantdisease/pathogen-articles/pathogens-common-many-plants/bacteria-other-prokaryotes/diseases (accessed on 22 May 2022).
9. Donati, I.; Cellini, A.; Sangiorgio, D.; Vanneste, J.L.; Scortichini, M.; Balestra, G.M.; Spinelli, F. *Pseudomonas syringae* pv. *actinidiae*: Ecology, Infection Dynamics and Disease Epidemiology. *Microb. Ecol.* **2020**, *80*, 81–102. [CrossRef]
10. Cameron, A.; Sarojini, V. *Pseudomonas syringae* pv. *actinidiae*: Chemical Control, Resistance Mechanisms and Possible Alternatives. *Plant Pathol.* **2014**, *63*, 1–11. [CrossRef]
11. Krasnovskaya, O.; Naumov, A.; Guk, D.; Gorelkin, P.; Erofeev, A.; Beloglazkina, E.; Majouga, A. Copper Coordination Compounds as Biologically Active Agents. *Int. J. Mol. Sci.* **2020**, *21*, 3965. [CrossRef] [PubMed]
12. Baker-Austin, C.; Wright, M.S.; Stepanauskas, R.; McArthur, J.V. Co-Selection of Antibiotic and Metal Resistance. *Trends Microbiol.* **2006**, *14*, 176–182. [CrossRef]
13. Seiler, C.; Berendonk, T.U. Heavy Metal Driven Co-Selection of Antibiotic Resistance in Soil and Water Bodies Impacted by Agriculture and Aquaculture. *Front. Microbiol.* **2012**, *3*, 399. [CrossRef]
14. Dy, R.L.; Rigano, L.A.; Fineran, P.C. Phage-Based Biocontrol Strategies and Their Application in Agriculture and Aquaculture. *Biochem. Soc. Trans.* **2018**, *46*, 1605–1613. [CrossRef]
15. Svircev, A.; Roach, D.; Castle, A. Framing the Future with Bacteriophages in Agriculture. *Viruses* **2018**, *10*, 218. [CrossRef]
16. Eilenberg, J.; Hajek, A.; Lomer, C. Suggestions for Unifying the Terminology in Biological Control. *BioControl* **2001**, *46*, 387–400. [CrossRef]
17. Jones, J.B.; Vallad, G.E.; Iriarte, F.B.; Obradović, A.; Wernsing, M.H.; Jackson, L.E.; Balogh, B.; Hong, J.C.; Momol, M.T. Considerations for Using Bacteriophages for Plant Disease Control. *Bacteriophage* **2012**, *2*, e23857. [CrossRef]
18. ANASAC Baciforte. Available online: https://www.anasac.cl/agropecuario/productos/baciforte/ (accessed on 31 May 2023).
19. Bionativa Nacillus. Available online: http://www.bionativa.cl/web/productos/nacillus-pro-wp/#toggle-id-3 (accessed on 31 May 2023).
20. BAYER Serenade®Max. Available online: https://www.cropscience.bayer.es/Hidden-Pages/Productos/Biologicos/Serenade-Max (accessed on 31 May 2023).
21. James, S.L.; Rabiey, M.; Neuman, B.W.; Percival, G.; Jackson, R.W. Isolation, Characterization and Experimental Evolution of Phage That Infect the Horse Chestnut Tree Pathogen, *Pseudomonas syringae* pv. *aesculi. Curr. Microbiol.* **2020**, *77*, 1438–1447. [CrossRef]
22. Cuppels, D.A. Isolation and Characterization of Phages Useful for Identifying *Pseudomonas syringae* pv. *tomato*. *Phytopathology* **1983**, *73*, 1376. [CrossRef]
23. Frampton, R.A.; Taylor, C.; Holguín Moreno, A.v.; Visnovsky, S.B.; Petty, N.K.; Pitman, A.R.; Fineran, P.C. Identification of Bacteriophages for Biocontrol of the Kiwifruit Canker Phytopathogen *Pseudomonas syringae* pv. *actinidiae*. *Appl. Environ. Microbiol.* **2014**, *80*, 2216–2228. [CrossRef] [PubMed]
24. Rombouts, S.; Volckaert, A.; Venneman, S.; Declercq, B.; Vandenheuvel, D.; Allonsius, C.N.; van Malderghem, C.; Jang, H.B.; Briers, Y.; Noben, J.P.; et al. Characterization of Novel Bacteriophages for Biocontrol of Bacterial Blight in Leek Caused by *Pseudomonas syringae* pv. *porri*. *Front. Microbiol.* **2016**, *7*, 279. [CrossRef] [PubMed]
25. Amarillas, L.; Estrada-Acosta, M.; León-Chan, R.G.; López-Orona, C.; Lightbourn, L. Complete Genome Sequence of Phobos: A Novel Bacteriophage with Unusual Genomic Features That Infects *Pseudomonas syringae*. *Arch. Virol.* **2020**, *165*, 1485–1488. [CrossRef]
26. Flores, O.; Retamales, J.; Núñez, M.; León, M.; Salinas, P.; Besoain, X.; Yañez, C.; Bastías, R. Characterization of Bacteriophages against *Pseudomonas syringae* pv. *actinidiae* with Potential Use as Natural Antimicrobials in Kiwifruit Plants. *Microorganisms* **2020**, *8*, 974. [CrossRef]
27. van Hall Pathogenicity and Identification of the Lilac Pathogen, *Pseudomonas syringae* pv. *syringae. Ann. Appl. Biol.* **1992**, *118*, 283–298. [CrossRef]
28. Morris, C.E.; Sands, D.C.; Vinatzer, B.A.; Glaux, C.; Guilbaud, C.; Buffière, A.; Yan, S.; Dominguez, H.; Thompson, B.M. The Life History of the Plant Pathogen *Pseudomonas syringae* Is Linked to the Water Cycle. *ISME J.* **2008**, *2*, 321–334. [CrossRef] [PubMed]
29. Stapp, C. *Handbuchdel Pflanzenkrank-Heiten. II Diepflanzlichen Parasiten*; Sorauer, P., Ed.; Paul Parey: Berlin, Germany, 1928.
30. Burkholder, W.H. The Bacterial Diseases of the Bean. *Memoirs. Cornell Univ. Agric. Exp. Stn.* **1930**, *127*, 1–88.
31. Robbs, C.F. Uma Nova Doença Bacteriana Do Mamoeiro (*Carica papaya* L.). *Rev. Soc. Bras. Agron.* **1956**, *12*, 73–76.
32. Psallidas, P.G.; Pamagopoulos, C.G. New Bacteriosis of Almond Caused by *Pseudomonas amygdali* sp. nov. *Annales* **1975**, *11*, 94–108.

33. Ogimi, C. Studies on Bacterial Gall of Chinaberry (*Melia Azedarach* Lin.), Caused by *Pseudomonas meliae* n. sp. *Sci. Bull. Coll. Agric. Univ. Ryukyu Jpn.* **1977**, *24*, 497–556.
34. Goto, M. *Pseudomonas ficuserectae* sp. nov., the Causal Agent of Bacterial Leaf Spot of *Ficus erecta* Thunb. *Int. J. Syst. Evol. Microbiol.* **1983**, *33*, 546–550. [CrossRef]
35. Janse, J.D.; Rossi, P.; Angelucci, L.; Scortichini, M.; Derks, J.H.J.; Akkermans, A.D.L.; De Vrijer, R.; Psallidas, P.G. Reclassification of *Pseudomonas syringae* pv. *avellanae* as *Pseudomonas avellanae* (spec. nov, the Bacterium Causing Canker of Hazelnut (*Corylus avellana* L.). *Syst. Appl. Microbiol.* **1996**, *19*, 589–595. [CrossRef]
36. Cardan, L.; Shafik, H.; Belouin, S.; Broch, R.; Grimont, F.; Grimont, P.A.D. DNA Relatedness among the Pathovars of *Pseudomonas syringae* and Description of *Pseudomonas tremae* sp. nov. and *Pseudomonas cannabina* sp. nov. (Ex Sutic and Dowson 1959). *Int. J. Syst. Bacteriol.* **1999**, *49*, 469–478. [CrossRef]
37. Behrendt, U.; Ulrich, A.; Schumann, P. Fluorescent Pseudomonads Associated with the Phyllosphere of Grasses; *Pseudomonas trivialis* sp. nov., *Pseudomonas poae* sp. nov. and *Pseudomonas congelans* sp. nov.. *Int. J. Syst. Evol. Microbiol.* **2003**, *53*, 1461–1469. [CrossRef]
38. González, A.J.; Cleenwerck, I.; De Vos, P.; Fernández-Sanz, A.M. *Pseudomonas asturiensis* sp. nov., Isolated from Soybean and Weeds. *Syst. Appl. Microbiol.* **2013**, *36*, 320–324. [CrossRef] [PubMed]
39. Kałużna, M.; Willems, A.; Pothier, J.F.; Ruinelli, M.; Sobiczewski, P.; Puławska, J. *Pseudomonas cerasi* sp. nov. (Non Griffin, 1911) Isolated from Diseased Tissue of Cherry. *Syst. Appl. Microbiol.* **2016**, *39*, 370–377. [CrossRef] [PubMed]
40. Busquets, A.; Gomila, M.; Beiki, F.; Mulet, M.; Rahimian, H.; García-Valdés, E.; Lalucat, J. *Pseudomonas caspiana* sp. nov., a Citrus Pathogen in the *Pseudomonas syringae* Phylogenetic Group. *Syst. Appl. Microbiol.* **2017**, *40*, 266–273. [CrossRef] [PubMed]
41. Schaad, N.W.; Cunfer, B.M. Synonymy of *Pseudomonas coronafaciens*, *Pseudomonas coronafaciens* Pathovar *zeae*, *Pseudomonas coronafaciens* subsp. *atropurpurea* and *Pseudomonas striafaciens*. *Int. J. Syst. Evol. Microbiol.* **1979**, *29*, 213–221. [CrossRef]
42. Gomila, M.; Busquets, A.; Mulet, M.; García-Valdés, E.; Lalucat, J. Clarification of Taxonomic Status within the *Pseudomonas syringae* Species Group Based on a Phylogenomic Analysis. *Front. Microbiol.* **2017**, *8*, 2422. [CrossRef]
43. Bull, C.T.; de Boer, S.H.; Denny, T.P.; Firrao, G.; Fischer-Le Saux, M.; Saddler, G.S.; Scortichini, M.; Stead, D.E.; Takikawa, Y. Comprehensive list of names of plant pathogenic bacteria, 1980–2007. *J. Plant Pathol.* **2010**, *92*, 551–592.
44. Berge, O.; Monteil, C.L.; Bartoli, C.; Chandeysson, C.; Guilbaud, C.; Sands, D.C.; Morris, C.E. A User's Guide to a Data Base of the Diversity of *Pseudomonas syringae* and Its Application to Classifying Strains in This Phylogenetic Complex. *PLoS ONE* **2014**, *9*, e105547. [CrossRef]
45. Pseudomonas syringae (Bacterial Blast). In *PlantwisePlus Knowledge Bank 2022*; CABI International: Wallingford, UK, 2022. [CrossRef]
46. Lamichhane, J.R.; Varvaro, L.; Parisi, L.; Audergon, J.-M.M.; Morris, C.E. Disease and Frost Damage of Woody Plants Caused by *Pseudomonas syringae*: Seeing the Forest for the Trees. *Adv. Agron.* **2014**, *126*, 235–295.
47. Lamichhane, J.R.; Messéan, A.; Morris, C.E. Insights into Epidemiology and Control of Diseases of Annual Plants Caused by the *Pseudomonas syringae* Species Complex. *J. Gen. Plant Pathol.* **2015**, *81*, 331–350. [CrossRef]
48. Hulin, M.T.; Mansfield, J.W.; Brain, P.; Xu, X.; Jackson, R.W.; Harrison, R.J. Characterization of the Pathogenicity of Strains of *Pseudomonas syringae* towards Cherry and Plum. *Plant Pathol.* **2018**, *67*, 1177–1193. [CrossRef]
49. Bultreys, A.; Kaluzna, M. Minerview Bacterial Cankers Caused by (*Pseudomonas syringae*) on Stone Fruit Species with Special Emphasis on the (Pathovars *syringae*) and (*morsprunorum*) Race 1 and Race 2. *J. Plant Pathol.* **2010**, *92*, S21–S33.
50. Ivanović, Ž.; Perović, T.; Popović, T.; Blagojević, J.; Trkulja, N.; Hrnčić, S. Characterization of *Pseudomonas syringae* pv. *syringae*, Causal Agent of Citrus Blast of Mandarin in Montenegro. *Plant Pathol. J.* **2017**, *33*, 21–33. [CrossRef]
51. Aiello, D.; Ferrante, P.; Vitale, A.; Polizzi, G.; Scortichini, M.; Cirvilleri, G. Characterization of *Pseudomonas syringae* pv. *syringae* Isolated from Mango in Sicily and Occurrence of Copper-Resistant Strains. *J. Plant Pathol.* **2015**, *97*, 273–282. [CrossRef]
52. Naqvi, S.A.H.; Perveen, R.; Rehman, A.U.; Khan, T.; Malik, M.T.; Chohan, S.; Tariq, A.; Akram, S.; Abbas, S.H. Outbreak of Bacterial Apical Necrosis of Mango in Multan, Punjab, Pakistan. *Pak. J. Phytopathol.* **2016**, *28*, 107–113.
53. Mazzaglia, A.; Studholme, D.J.; Taratufolo, M.C.; Cai, R.; Almeida, N.F.; Goodman, T.; Guttman, D.S.; Vinatzer, B.A.; Balestra, G.M. *Pseudomonas syringae* pv. *actinidiae* (*Psa*) Isolates from Recent Bacterial Canker of Kiwifruit Outbreaks Belong to the Same Genetic Lineage. *PLoS ONE* **2012**, *7*, e36518. [CrossRef]
54. Scortichini, M.; Marcelletti, S.; Ferrante, P.; Petriccione, M.; Firrao, G. *Pseudomonas syringae* pv. *actinidiae*: A Re-Emerging, Multi-Faceted, Pandemic Pathogen. *Mol. Plant Pathol.* **2012**, *13*, 631–640. [CrossRef]
55. Cameron, A.; De Zoysa, G.H.; Sarojini, V. Antimicrobial Peptides against *Pseudomonas syringae* pv. *actinidiae* and *Erwinia amylovora*: Chemical Synthesis, Secondary Structure, Efficacy, and Mechanistic Investigations. *Biopolymers* **2014**, *102*, 88–96. [CrossRef] [PubMed]
56. Gallelli, A.; Talocci, S.; Pilotti, M.; Loreti, S. Real-Time and Qualitative PCR for Detecting *Pseudomonas syringae* pv. *actinidiae* Isolates Causing Recent Outbreaks of Kiwifruit Bacterial Canker. *Plant Pathol.* **2014**, *63*, 264–276. [CrossRef]
57. Vanneste, J.L.; Reglinski, T.; Yu, J.; Cornish, D.A. Multiplication and Movement of *Pseudomonas syringae* pv. *actinidiae* in Kiwifruit Plants. *Acta Hortic.* **2015**, *1095*, 117–122. [CrossRef]
58. Prencipe, S.; Gullino, M.L.; Spadaro, D. *Pseudomonas syringae* pv. *actinidiae* Isolated from *Actinidia chinensis* Var. *Deliciosa* in Northern Italy: Genetic Diversity and Virulence. *Eur. J. Plant Pathol.* **2018**, *150*, 191–204. [CrossRef]

59. Ferrante, P.; Scortichini, M. Redefining the Global Populations of *Pseudomonas syringae* pv. *actinidiae* Based on Pathogenic, Molecular and Phenotypic Characteristics. *Plant Pathol.* **2015**, *64*, 51–62. [CrossRef]
60. Morris, C.E.; Monteil, C.L.; Berge, O. The Life History of *Pseudomonas syringae*: Linking Agriculture to Earth System Processes. *Annu. Rev. Phytopathol.* **2013**, *51*, 85–104. [CrossRef] [PubMed]
61. Basim, H.; Basim, E.; Yilmaz, S.; Dickstein, E.R.; Jones, J.B. An Outbreak of Bacterial Speck Caused by *Pseudomonas syringae* pv. *tomato* on Tomato Transplants Grown in Commercial Seedling Companies Located in the Western Mediterranean Region of Turkey. *Plant Dis.* **2004**, *88*, 1050. [CrossRef]
62. Cruz, L.; Cruz, J.; Eloy, M.; Oliveira, H.; Vaz, H.; Tenreiro, R. First Report of Bacterial Speck of Tomato Caused by *Pseudomonas syringae* pv. *tomato* Race 1 in Portugal. *Plant Dis.* **2010**, *94*, 1504. [CrossRef]
63. Takikawa, Y.; Takahashi, F. Bacterial Leaf Spot and Blight of Crucifer Plants (Brassicaceae) Caused by *Pseudomonas syringae* pv. *maculicola* and *P. cannabina* pv. *alisalensis*. *J. Gen. Plant Pathol.* **2014**, *80*, 466–474. [CrossRef]
64. Shila, S.J.; Islam, M.R.; Ahmed, N.N.; Dastogeer, K.M.G.; Meah, M.B. Detection of *Pseudomonas syringae* pv. *lachrymans* Associated with the Seeds of Cucurbits. *Univ. J. Agric. Res.* **2013**, *1*, 1–8. [CrossRef]
65. Newberry, E.A.; Babu, B.; Roberts, P.D.; Dufault, N.S.; Goss, E.M.; Jones, J.B.; Paret, M.L. Molecular Epidemiology of *Pseudomonas syringae* pv. *syringae* Causing Bacterial Leaf Spot of Watermelon and Squash in Florida. *Plant Dis.* **2018**, *102*, 511–518. [CrossRef]
66. Martín-Sanz, A.; Pérez de la Vega, M.; Murillo, J.; Caminero, C. Genetic, Biochemical and Pathogenic Diversity of *Pseudomonas syringae* pv. *pisi* Strains. *Plant Pathol.* **2012**, *61*, 1063–1072. [CrossRef]
67. Xin, X.F.; Kvitko, B.; He, S.Y. *Pseudomonas syringae*: What It Takes to Be a Pathogen. *Nat. Rev. Microbiol.* **2018**, *16*, 316–328. [CrossRef]
68. Pscheidt, J.W.; Warneke, B.W.; Nackley, L.L. Efficacy of tank mixing biological fungicides with sulfur for management of grape powdery mildew. GRAPE (Vitis Vinifera 'Chardonnay'). 2022.
69. Hinrichs-Berger, J. Epidemiology of *Pseudomonas syringae* Pathovars Associated with Decline of Plum Trees in the Southwest of Germany. *J. Phytopathol.* **2004**, *152*, 153–160. [CrossRef]
70. Iličić, R.; Balaž, J.; Ognjanov, V.; Popović, T. Epidemiology Studies of *Pseudomonas syringae* Pathovars Associated with Bacterial Canker on the Sweet Cherry in Serbia. *Plant Prot. Sci.* **2021**, *57*, 196–205. [CrossRef]
71. Umiraliyeva, Z.Z.; Kopzhassarov, B.K.; Jaimurzina, A.A.; Niyazbekov, Z.B.; Issenova, G.Z.; Tursunova, A.K.; Berganayeva, G.E. Epidemiology of Fire Blight in Fruit Crops in Kazakhstan. *Agrivita* **2021**, *43*, 273–284. [CrossRef]
72. Mauri, S.; Cellini, A.; Buriani, G.; Donati, I.; Costa, G.; Spinelli, F. Optimization of Cultural Practices to Reduce the Development of *Pseudomonas syringae* pv. *actinidiae*, Causal Agent of the Bacterial Canker of Kiwifruit. *J. Berry Res.* **2016**, *6*, 355–371. [CrossRef]
73. Vanneste, J.L.; Poliakoff, F.; Audusseau, C.; Cornish, D.A.; Paillard, S.; Rivoal, C.; Yu, J. First Report of *Pseudomonas syringae* pv. *actinidiae*, the Causal Agent of Bacterial Canker of Kiwifruit in France. *Plant Dis.* **2011**, *95*, 1311. [CrossRef] [PubMed]
74. Lee, J.H.; Kim, J.H.; Kim, G.H.; Jung, J.S.; Hur, J.S.; Koh, Y.J. Comparative Analysis of Korean and Japanese Strains of *Pseudomonas syringae* pv. *actinidiae* Causing Bacterial Canker of Kiwifruit. *Plant Pathol. J.* **2005**, *21*, 119–126. [CrossRef]
75. Altimira, F.; Yáñez, C.; Bravo, G.; González, M.; Rojas, L.A.; Seeger, M. Characterization of Copper-Resistant Bacteria and Bacterial Communities from Copper-Polluted Agricultural Soils of Central Chile. *BMC Microbiol.* **2012**, *12*, 193. [CrossRef] [PubMed]
76. Jones, J.B.; Jackson, L.E.; Balogh, B.; Obradovic, A.; Iriarte, F.B.; Momol, M.T. Bacteriophages for Plant Disease Control. *Annu. Rev. Phytopathol.* **2007**, *45*, 245–262. [CrossRef] [PubMed]
77. Lamichhane, J.R.; Osdaghi, E.; Behlau, F.; Köhl, J.; Jones, J.B.; Aubertot, J.N. Thirteen Decades of Antimicrobial Copper Compounds Applied in Agriculture. A Review. *Agron. Sustain. Dev.* **2018**, *38*, 28. [CrossRef]
78. Pinheiro, L.A.M.; Pereira, C.; Frazão, C.; Balcão, V.M.; Almeida, A. Efficiency of Phage Φ6 for Biocontrol of *Pseudomonas syringae* pv. *syringae*: An in Vitro Preliminary Study. *Microorganisms* **2019**, *7*, 286. [CrossRef]
79. Frampton, R.A.; Pitman, A.R.; Fineran, P.C. Advances in Bacteriophage-Mediated Control of Plant Pathogens. *Int. J. Microbiol.* **2012**, *2012*, 326452. [CrossRef]
80. Kering, K.K.; Kibii, B.J.; Wei, H. Biocontrol of Phytobacteria with Bacteriophage Cocktails. *Pest Manag. Sci.* **2019**, *75*, 1775–1781. [CrossRef] [PubMed]
81. Fagnano, M.; Agrelli, D.; Pascale, A.; Adamo, P.; Fiorentino, N.; Rocco, C.; Pepe, O.; Ventorino, V. Copper Accumulation in Agricultural Soils: Risks for the Food Chain and Soil Microbial Populations. *Sci. Total Environ.* **2020**, *734*, 139434. [CrossRef] [PubMed]
82. Coelho, F.C.; Squitti, R.; Ventriglia, M.; Cerchiaro, G.; Daher, J.P.; Rocha, J.G.; Rongioletti, M.C.A.; Moonen, A.-C. Biomolecules Agricultural Use of Copper and Its Link to Alzheimer's Disease. *Biomolecules* **2020**, *10*, 897. [CrossRef] [PubMed]
83. Knowles, A. Recent Developments of Safer Formulations of Agrochemicals. *Environmentalist* **2008**, *28*, 35–44. [CrossRef]
84. Czajkowski, R.; Ozymko, Z.; Lojkowska, E. Isolation and Characterization of Novel Soilborne Lytic Bacteriophages Infecting *Dickeya* Spp. Biovar 3 ('*D. Solani*'). *Plant Pathol.* **2013**, *63*, 758–772. [CrossRef]
85. Conlin, K.C. Effectiveness of Selected Chemicals in Inhibiting *Pseudomonas syringae* pv. *tomato* in vitro and in Controlling Bacterial Speck. *Plant Dis.* **1983**, *67*, 639–644. [CrossRef]
86. McLeod, A.; Masimba, T.; Jensen, T.; Serfontein, K.; Coertze, S. Evaluating Spray Programs for Managing Copper Resistant *Pseudomonas syringae* pv. *tomato* Populations on Tomato in the Limpopo Region of South Africa. *Crop Prot.* **2017**, *102*, 32–42. [CrossRef]

87. Wilson, M.; Campbell, H.L.; Ji, P.; Jones, J.B.; Cuppels, D.A. Biological Control of Bacterial Speck of Tomato under Field Conditions at Several Locations in North America. *Phytopathology* **2002**, *92*, 1284–1292. [CrossRef]
88. Belpoggi, F.; Soffritti, M.; Guarino, M.; Lambertini, L.; Cevolani, D.; Maltoni, C. Results of Long-Term Experimental Studies on the Carcinogenicity of Ethylene-Bis-Dithiocarbamate (Mancozeb) in Rats. *Ann. N. Y. Acad. Sci.* **2002**, *982*, 123–136. [CrossRef]
89. Tripathi, D.; Raikhy, G.; Kumar, D. Chemical Elicitors of Systemic Acquired Resistance—Salicylic Acid and Its Functional Analogs. *Curr. Plant Biol.* **2019**, *17*, 48–59. [CrossRef]
90. Stroud, E.A.; Rikkerink, E.H.A.; Jayaraman, J.; Templeton, M.D. Actigard™ Induces a Defence Response to Limit *Pseudomonas syringae* pv. *actinidiae* in *Actinidia chinensis* var. *chinensis* 'Hort16A' Tissue Culture Plants. *Sci. Hortic.* **2022**, *295*, 110806. [CrossRef]
91. Wurms, K.V.; Gould, E.; Ah Chee, A.; Taylor, J.; Curran, B.; Reglinski, T. Elicitor Induction of Defense Genes and Reduction of Bacterial Canker in Kiwifruit. *N. Z. Plant Prot.* **2017**, *70*, 272–284. [CrossRef]
92. Michelotti, V.; Lamontanara, A.; Buriani, G.; Orrù, L.; Cellini, A.; Donati, I.; Vanneste, J.L.; Cattivelli, L.; Tacconi, G.; Spinelli, F. Comparative Transcriptome Analysis of the Interaction between *Actinidia chinensis* var. *chinensis* and *Pseudomonas syringae* pv. *actinidiae* in Absence and Presence of Acibenzolar-S-Methyl. *BMC Genom.* **2018**, *19*, 585. [CrossRef]
93. Stout, M.J.; Fidantsef, A.L.; Duffey, S.S.; Bostock, R.M. Signal Interactions in Pathogen and Insect Attack: Systemic Plant-Mediated Interactions between Pathogens and Herbivores of the Tomato, Lycopersicon Esculentum. *Physiol. Mol. Plant Pathol.* **1999**, *54*, 115–130. [CrossRef]
94. Reglinski, T.; Vanneste, J.L.; Wurms, K.; Gould, E.; Spinelli, F.; Rikkerink, E. Using Fundamental Knowledge of Induced Resistance to Develop Control Strategies for Bacterial Canker of Kiwifruit Caused by *Pseudomonas syringae* pv. *actinidiae*. *Front. Plant Sci.* **2013**, *4*, 24. [CrossRef] [PubMed]
95. Shafi, J.; Tian, H.; Ji, M. *Bacillus* Species as Versatile Weapons for Plant Pathogens: A Review. *Biotechnol. Biotechnol. Equip.* **2017**, *31*, 446–459. [CrossRef]
96. Hwang, M.S.H.; Morgan, R.L.; Sarkar, S.F.; Wang, P.W.; Guttman, D.S. Phylogenetic Characterization of Virulence and Resistance Phenotypes of *Pseudomonas syringae*. *Appl. Environ. Microbiol.* **2005**, *71*, 5182–5191. [CrossRef]
97. Sundin, G.W.; Wang, N. Antibiotic Resistance in Plant-Pathogenic Bacteria. *Annu. Rev. Phytopathol.* **2018**, *56*, 161–180. [CrossRef] [PubMed]
98. Cooksey, D.A. Molecular Mechanisms of Copper Resistance and Accumulation in Bacteria. *FEMS Microbiol. Rev.* **1994**, *14*, 381–386. [CrossRef] [PubMed]
99. Behlau, F.; Canteros, B.I.; Jones, J.B.; Graham, J.H. Copper Resistance Genes from Different *Xanthomonads* and Citrus Epiphytic Bacteria Confer Resistance to *Xanthomonas citri* subsp. *citri*. *Eur. J. Plant Pathol.* **2012**, *133*, 949–963. [CrossRef]
100. Xu, Y.; Luo, Q.; Zhou, M.G. Identification and Characterization of Integron-Mediated Antibiotic Resistance in the Phytopathogen *Xanthomonas oryzae* pv. *oryzae*. *PLoS ONE* **2013**, *8*, e55962. [CrossRef]
101. Argüello, J.M.; Raimunda, D.; Padilla-Benavides, T. Mechanisms of Copper Homeostasis in Bacteria. *Front. Cell. Infect. Microbiol.* **2013**, *4*, 73. [CrossRef]
102. Adaikkalam, V.; Swarup, S. Molecular Characterization of an Operon, CueAR, Encoding a Putative P1-Type ATPase and a MerR-Type Regulatory Protein Involved in Copper Homeostasis in *Pseudomonas putida*. *Microbiology* **2002**, *148*, 2857–2867. [CrossRef]
103. Merry, R.H.; Tiller, K.G.; Alston, A.M. Accumulation of Copper, Lead and Arsenic in Some Australian Orchard Soils. *Aust. J. Soil Res.* **1983**, *21*, 549–561. [CrossRef]
104. Griffin, K.; Campbell, P.; Gambley, C. Genetic Basis of Copper-Tolerance in Australian *Pseudomonas syringae* pv. *tomato*. *Australas. Plant Pathol.* **2019**, *48*, 425–437. [CrossRef]
105. Teixeira, E.C.; de Oliveira, J.C.F.; Novo, M.T.M.; Bertolini, M.C. The Copper Resistance Operon CopAB from *Xanthomonas axonopodis* Pathovar *citri*: Gene Inactivation Results in Copper Sensitivity. *Microbiology* **2008**, *154*, 402–412. [CrossRef]
106. Bondarczuk, K.; Piotrowska-Seget, Z. Molecular Basis of Active Copper Resistance Mechanisms in Gram-Negative Bacteria. *Cell Biol. Toxicol.* **2013**, *29*, 397–405. [CrossRef]
107. Bender, C.L.; Cooksey, D.A. Molecular Cloning of Copper Resistance Genes from *Pseudomonas syringae* pv. *tomato*. *J. Bacteriol.* **1987**, *169*, 470–474. [CrossRef] [PubMed]
108. Ma, Z.; Smith, J.J.; Zhao, Y.; Jackson, R.W.; Arnold, D.L.; Murillo, J.; Sundin, G.W. Phylogenetic Analysis of the PPT23A Plasmid Family of *Pseudomonas syringae*. *Appl. Environ. Microbiol.* **2007**, *73*, 1287–1295. [CrossRef]
109. Gutiérrez-Barranquero, J.A.; de Vicente, A.; Carrión, V.J.; Sundin, G.W.; Cazorla, F.M. Recruitment and Rearrangement of Three Different Genetic Determinants into a Conjugative Plasmid Increase Copper Resistance in *Pseudomonas syringae*. *Appl. Environ. Microbiol.* **2013**, *79*, 1028–1033. [CrossRef]
110. Colombi, E.; Straub, C.; Künzel, S.; Templeton, M.D.; McCann, H.C.; Rainey, P.B. Evolution of Copper Resistance in the Kiwifruit Pathogen *Pseudomonas syringae* pv. *actinidiae* through Acquisition of Integrative Conjugative Elements and Plasmids. *Environ. Microbiol.* **2017**, *19*, 819–832. [CrossRef]
111. Gutiérrez-Barranquero, J.A.; Cazorla, F.M.; de Vicente, A.; Sundin, G.W. Complete Sequence and Comparative Genomic Analysis of Eight Native *Pseudomonas syringae* Plasmids Belonging to the PPT23A Family. *BMC Genom.* **2017**, *18*, 365. [CrossRef]
112. Sundin, G.W.; Bender, C.L. Expression of the StrA-StrB Streptomycin Resistance Genes in *Pseudomonas syringae* and *Xanthomonas campestris* and Characterization of IS6100 in *X. campestris*. *Appl. Environ. Microbiol.* **1995**, *61*, 2891–2897. [CrossRef] [PubMed]

113. Han, H.; Nam, H.; Koh, Y.; Hur, J.; Jung, J. Molecular Bases of High-Level Streptomycin Resistance in *Pseudomonas marginalis* and *Pseudomonas syringae* pv. *actinidiae*. *J. Microbiol.* **2003**, *41*, 16–21.
114. Nakajima, M.; Yamashita, S.; Takikawa, Y.; Tsuyumu, S.; Hibi, T.; Goto, M. Similarity of Streptomycin Resistance Gene(s) in *Pseudomonas syringae* pv. *actinidiae* with StrA and StrB of Plasmid RSF1010. *Jpn. J. Phytopathol.* **1995**, *61*, 489–492. [CrossRef]
115. Collis, C.M.; Hall, R.M. Expression of Antibiotic Resistance Genes in the Integrated Cassettes of Integrons. *Antimicrob. Agents Chemother.* **1995**, *39*, 155–162. [CrossRef]
116. Marcelletti, S.; Ferrante, P.; Petriccione, M.; Firrao, G.; Scortichini, M. *Pseudomonas syringae* pv. *actinidiae* Draft Genomes Comparison Reveal Strain-Specific Features Involved in Adaptation and Virulence to Actinidia Species. *PLoS ONE* **2011**, *6*, e27297. [CrossRef]
117. Walsh, C. Molecular Mechanisms That Confer Antibacterial Drug Resistance. *Nature* **2000**, *406*, 775–781. [CrossRef]
118. Jacoby, G.A.; Archer, G.L. New Mechanisms of Bacterial Resistance to Antimicrobial Agents. *N. Engl. J. Med.* **1991**, *324*, 601–612. [CrossRef] [PubMed]
119. Vanneste, J.L.; Voyle, M.D.; Yu, J.; Cornish, D.A.; Boyd, R.J.; Mclaren, G.F. Copper and Streptomycin Resistance in *Pseudomonas* Strains Isolated from Pipfruit and Stone Fruit Orchards in New Zealand. In *Pseudomonas syringae Pathovars and Related Pathogens—Identification, Epidemiology and Genomics*; Springer: Berlin/Heidelberg, Germany, 2008; pp. 81–90. [CrossRef]
120. Shenge, K.C.; Wydra, K.; Mabagala, R.B.; Mortensen, C.N. Assessment of Strains of *Pseudomonas syringae* pv. *tomato* from Tanzania for Resistance to Copper and Streptomycin. *Arch. Phytopathol. Plant Prot.* **2008**, *41*, 572–585. [CrossRef]
121. Glibota, N.; Grande Burgos, M.J.; Gálvez, A.; Ortega, E. Copper Tolerance and Antibiotic Resistance in Soil Bacteria from Olive Tree Agricultural Fields Routinely Treated with Copper Compounds. *J. Sci. Food Agric.* **2019**, *99*, 4677–4685. [CrossRef]
122. Córdova, P.; Rivera-González, J.P.; Rojas-Martínez, V.; Villarreal, P.; Zamorano, A.; Fiore, N.; San Martín, D.; Vera, F.; Gálvez, E.; Romero, J.; et al. Antimicrobial Multiresistant Phenotypes of Genetically Diverse *Pseudomonas* spp. Isolates Associated with Tomato Plants in Chilean Orchards. *Horticulturae* **2022**, *8*, 750. [CrossRef]
123. Mann, A.; Nehra, K.; Rana, J.S.; Dahiya, T. Antibiotic Resistance in Agriculture: Perspectives on Upcoming Strategies to Overcome Upsurge in Resistance. *Curr. Res. Microb. Sci.* **2021**, *2*, 100030. [CrossRef]
124. Freeland, G.; Hettiarachchy, N.; Atungulu, G.G.; Apple, J.; Mukherjee, S. Strategies to Combat Antimicrobial Resistance from Farm to Table. *Food Rev. Int.* **2021**, *39*, 27–40. [CrossRef]
125. Murugaiyan, J.; Anand Kumar, P.; Rao, G.S.; Iskandar, K.; Hawser, S.; Hays, J.P.; Mohsen, Y.; Adukkadukkam, S.; Awuah, W.A.; Jose, R.A.M.; et al. Progress in Alternative Strategies to Combat Antimicrobial Resistance: Focus on Antibiotics. *Antibiotics* **2022**, *11*, 200. [CrossRef]
126. Stenberg, J.A. A Conceptual Framework for Integrated Pest Management. *Trends Plant Sci.* **2017**, *22*, 759–769. [CrossRef] [PubMed]
127. Caruana, J.C.; Dhar, N.; Raina, R. Overexpression of Arabidopsis MicroRNA167 Induces Salicylic Acid-Dependent Defense against *Pseudomonas syringae* through the Regulation of Its Targets ARF6 and ARF8. *Plant Direct* **2020**, *4*, e00270. [CrossRef] [PubMed]
128. Huang, Y.; Nordeen, R.O.; Di, M.; Owens, L.D.; McBeath, J.H. Expression of an Engineered Cecropin Gene Cassette in Transgenic Tobacco Plants Confers Disease Resistance to *Pseudomonas syringae* pv. *tabaci*. *Phytopathology* **1997**, *87*, 494–499. [CrossRef]
129. Rooney, W.M.; Grinter, R.W.; Correia, A.; Parkhill, J.; Walker, D.C.; Milner, J.J. Engineering Bacteriocin-Mediated Resistance against the Plant Pathogen *Pseudomonas syringae*. *Plant Biotechnol. J.* **2020**, *18*, 1296–1306. [CrossRef]
130. Téllez, J.; Muñoz-Barrios, A.; Sopeña-Torres, S.; Martín-Forero, A.F.; Ortega, A.; Pérez, R.; Sanz, Y.; Borja, M.; de Marcos, A.; Nicolas, M.; et al. YODA Kinase Controls a Novel Immune Pathway of Tomato Conferring Enhanced Disease Resistance to the Bacterium *Pseudomonas syringae*. *Front. Plant Sci.* **2020**, *11*, 584471. [CrossRef] [PubMed]
131. Xu, P.; Ma, W.; Liu, J.; Hu, J.; Cai, W. Overexpression of a Small GTP-Binding Protein Ran1 in Arabidopsis Leads to Promoted Elongation Growth and Enhanced Disease Resistance against *P. syringae DC*3000. *Plant J.* **2021**, *108*, 977–991. [CrossRef]
132. Köhl, J.; Medeiros, F.H.V.; Lombaers-van der Plas, C.; Groenenboom-de Haas, L.; van den Bosch, T. Efficacies of Bacterial and Fungal Isolates in Biocontrol of *Botrytis cinerea* and *Pseudomonas syringae* pv. *tomato* and Growth Promotion in Tomato Do Not Correlate. *Biol. Control* **2020**, *150*, 104375. [CrossRef]
133. El-Wakeil, N.; Saleh, M.; Abu-hashim, M. Conclusions and Recommendations of Biological Control Industry. In *Cottage Industry of Biocontrol Agents and Their Applications: Practical Aspects to Deal Biologically with Pests and Stresses Facing Strategic Crops*; Springer: Cham, Switzerland, 2019; pp. 451–466.
134. Rabiey, M.; Roy, S.R.; Holtappels, D.; Franceschetti, L.; Quilty, B.J.; Creeth, R.; Sundin, G.W.; Wagemans, J.; Lavigne, R.; Jackson, R.W. Phage Biocontrol to Combat *Pseudomonas syringae* Pathogens Causing Disease in Cherry. *Microb. Biotechnol.* **2020**, *13*, 1428–1445. [CrossRef] [PubMed]
135. OmniLytics The Phage Company. Available online: https://www.omnilytics.com/ (accessed on 15 April 2023).
136. Biogard Amylo-X®. Available online: https://www.biogard.it/prodotto/amylo-x/ (accessed on 15 April 2023).
137. Mougou, I.; Boughalleb-M'hamdi, N. Biocontrol of *Pseudomonas syringae* pv. *syringae* Affecting Citrus Orchards in Tunisia by Using Indigenous *Bacillus* Spp. and Garlic Extract. *Egypt. J. Biol. Pest Control* **2018**, *28*, 60. [CrossRef]
138. Bais, H.P.; Fall, R.; Vivanco, J.M. Biocontrol of *Bacillus subtilis* against Infection of Arabidopsis Roots by *Pseudomonas syringae* Is Facilitated by Biofilm Formation and Surfactin Production. *Plant Physiol.* **2004**, *134*, 307–319. [CrossRef]
139. Braun-Kiewnick, A.; Jacobsen, B.J.; Sands, D.C. Biological Control of *Pseudomonas syringae* pv. *syringae*, the Causal Agent of Basal Kernel Blight of Barley, by Antagonistic *Pantoea agglomerans*. *Phytopathology* **2000**, *90*, 368–375. [CrossRef]

140. NuFarm Americas BlightBan®A506-US-Crop. Available online: https://nufarm.com/uscrop/product/blightbana506/ (accessed on 17 April 2023).
141. Hossain, M.M.; Sultana, F.; Kubota, M.; Koyama, H.; Hyakumachi, M. The Plant Growth-Promoting Fungus *Penicillium simplicissimum* GP17-2 Induces Resistance in *Arabidopsis thaliana* by Activation of Multiple Defense Signals. *Plant Cell Physiol.* **2007**, *48*, 1724–1736. [CrossRef]
142. Yoo, S.J.; Shin, D.J.; Won, H.Y.; Song, J.; Sang, M.K. *Aspergillus terreus* JF27 Promotes the Growth of Tomato Plants and Induces Resistance against *Pseudomonas syringae* pv. *tomato*. *Mycobiology* **2018**, *46*, 147–153. [CrossRef]
143. Heil, M.; Bostock, R.M. Induced Systemic Resistance (ISR) against Pathogens in the Context of Induced Plant Defences. *Ann. Bot.* **2002**, *89*, 503–512. [CrossRef]
144. Schneider, M.; Schweizer, P.; Meuwly, P.; Métraux, J.P. Systemic Acquired Resistance in Plants. *Plant Cell* **1996**, *168*, 303–340.
145. Botrel, D.A.; Laborde, M.C.F.; De Medeiros, F.H.V.; De Resende, M.L.V.; Ribeiro Júnio, P.M.; Pascholati, S.F.; Gusmão, L.F.P. Saprobic Fungi as Biocontrol Agents of Halo Blight (*Pseudomonas syringae* pv. *garcae*) in Coffee Clones. *Coffee Sci.* **2018**, *13*, 283–291. [CrossRef]
146. Hawksworth, D.L. The Fungal Dimension of Biodiversity: Magnitude, Significance, and Conservation. *Mycol. Res.* **1991**, *95*, 641–655. [CrossRef]
147. Schmit, J.P.; Mueller, G.M. An Estimate of the Lower Limit of Global Fungal Diversity. *Biodivers. Conserv.* **2007**, *16*, 99–111. [CrossRef]
148. Shoda, M. Bacterial Control of Plant Diseases. *J. Biosci. Bioeng.* **2000**, *89*, 515–521. [CrossRef]
149. Bashan, Y.; De-Bashan, L.E. Protection of Tomato Seedlings against Infection by *Pseudomonas syringae* pv. *tomato* by Using the Plant Growth-Promoting Bacterium *Azospirillum brasilense*. *Appl. Environ. Microbiol.* **2002**, *68*, 2637–2643. [CrossRef]
150. Ji, P.; Campbell, H.L.; Kloepper, J.W.; Jones, J.B.; Suslow, T.V.; Wilson, M. Integrated Biological Control of Bacterial Speck and Spot of Tomato under Field Conditions Using Foliar Biological Control Agents and Plant Growth-Promoting Rhizobacteria. *Biol. Control* **2006**, *36*, 358–367. [CrossRef]
151. Köhl, J.; Kolnaar, R.; Ravensberg, W.J. Mode of Action of Microbial Biological Control Agents against Plant Diseases: Relevance beyond Efficacy. *Front. Plant Sci.* **2019**, *10*, 845. [CrossRef] [PubMed]
152. Daranas, N.; Roselló, G.; Cabrefiga, J.; Donati, I.; Francés, J.; Badosa, E.; Spinelli, F.; Montesinos, E.; Bonaterra, A. Biological Control of Bacterial Plant Diseases with *Lactobacillus plantarum* Strains Selected for Their Broad-Spectrum Activity. *Ann. Appl. Biol.* **2019**, *174*, 92–105. [CrossRef] [PubMed]
153. Patel, H.; Tscheka, C.; Edwards, K.; Karlsson, G.; Heerklotz, H. All-or-None Membrane Permeabilization by Fengycin-Type Lipopeptides from *Bacillus subtilis* QST713. *Biochim. Biophys. Acta* **2011**, *1808*, 2000–2008. [CrossRef] [PubMed]
154. Wicaksono, W.A.; Jones, E.E.; Casonato, S.; Monk, J.; Ridgway, H.J. Biological Control of *Pseudomonas syringae* pv. *actinidiae* (Psa), the Causal Agent of Bacterial Canker of Kiwifruit, Using Endophytic Bacteria Recovered from a Medicinal Plant. *Biol. Control* **2018**, *116*, 103–112. [CrossRef]
155. Gentili, A.; Mariotti, E.; Vincenzi, A.; Mazzaglia, A.; Heydari, A.; Schaad, N.W.; Varvaro, L.; Balestra, G.M. Dieback (Moria) of Hazelnut: Isolation and Characterization of Two Potential Biocontrol Agents. *J. Plant Pathol.* **2009**, *90*, 383–386.
156. Großkinsky, D.K.; Tafner, R.; Moreno, M.V.; Stenglein, S.A.; De Salamone, I.E.G.; Nelson, L.M.; Novák, O.; Strnad, M.; Van Der Graaff, E.; Roitsch, T. Cytokinin Production by *Pseudomonas fluorescens* G20-18 Determines Biocontrol Activity against *Pseudomonas syringae* in Arabidopsis. *Sci. Rep.* **2016**, *6*, 23310. [CrossRef] [PubMed]
157. Dutta, S.; Yu, S.M.; Lee, Y.H. Assessment of the Contribution of Antagonistic Secondary Metabolites to the Antifungal and Biocontrol Activities of *Pseudomonas fluorescens* NBC275. *Plant Pathol. J.* **2020**, *36*, 491–496. [CrossRef]
158. Attia Abo-Zaid, G.; Abdel-Mohsen Soliman, N.; Salah Abdullah, A.; Ebrahim El-Sharouny, E.; Mohamed Matar, S.; Abdel-Fattah Sabry, S. Maximization of Siderophores Production from Biocontrol Agents, *Pseudomonas aeruginosa* F2 and *Pseudomonas fluorescens* JY3 Using Batch and Exponential Fed-Batch Fermentation. *Processes* **2020**, *8*, 455. [CrossRef]
159. Morella, N.M.; Zhang, X.; Koskella, B. Tomato Seed-Associated Bacteria Confer Protection of Seedlings against Foliar Disease Caused by *Pseudomonas syringae*. *Phytobiomes J.* **2019**, *3*, 177–190. [CrossRef]
160. Akbaba, M.; Ozaktan, H. Biocontrol of Angular Leaf Spot Disease and Colonization of Cucumber (*Cucumis sativus* L.) by Endophytic Bacteria. *Egypt. J. Biol. Pest Control* **2018**, *28*, 14. [CrossRef]
161. Atterbury, R.J.; Barrow, P.A. The Use of Bacteriophages in Veterinary Therapy. In *Bacteriophages*; Springer: Cham, Switzerland, 2021; pp. 953–987. [CrossRef]
162. Kotan, R.; Şahin, F. Biological Control of *Pseudomonas syringae* pv. *syringae* and Nutritional Similarity in Carbon Source Utilization of Pathogen and Its Potential Biocontrol Agents. *J. Turk. Phytopathol.* **2006**, *35*, 1–13.
163. Raveau, R.; Fontaine, J.; Lounès-Hadj Sahraoui, A. Essential Oils as Potential Alternative Biocontrol Products against Plant Pathogens and Weeds: A Review. *Foods* **2020**, *9*, 365. [CrossRef]
164. Fousia, S.; Paplomatas, E.J.; Tjamos, S.E. *Bacillus subtilis* QST 713 Confers Protection to Tomato Plants Against *Pseudomonas syringae* pv. *tomato* and Induces Plant Defence-Related Genes. *J. Phytopathol.* **2016**, *164*, 264–270. [CrossRef]
165. Völksch, B.; May, R. Biological Control of *Pseudomonas syringae* pv. *glycinea* by Epiphytic Bacteria under Field Conditions. *Microb. Ecol.* **2001**, *41*, 132–139. [CrossRef] [PubMed]
166. Suttle, C. Crystal Ball. The Viriosphere: The Greatest Biological Diversity on Earth and Driver of Global Processes. *Environ. Microbiol.* **2005**, *7*, 481–482. [CrossRef]

167. Fauquet, C.M.; Mayo, M.A.; Maniloff, J.; Desselberger, U.; Ball, L.A. *Virus Taxonomy: VIIIth Report of the International Committee on Taxonomy of Viruses*; Elsevier Science: Amsterdam, The Netherlands, 2005; ISBN 9780080575483.
168. Summers, W.C. Bacteriophage Therapy. *Annu. Rev. Microbiol.* **2001**, *55*, 437–451. [CrossRef]
169. Mallmann, W.L.; Hemstreet, C. Isolation of an Inhibitory Substance from Plants. *Agric. Res.* **1924**, *28*, 599–602.
170. Coons, G.H. The Transmissible Lytic Principle (Bacteriophage) in Relation to Plant Pathogens. *Phytopathology* **1925**, *15*, 357–370.
171. Kotila, J.E. *Investigations of the Blackleg Disease of the Potato*; Michigan Agricultural College: East Lansing, MI, USA, 1925; Volume 67, pp. 3–29.
172. Thomas, R.C. A Bacteriophage in Relation to Stewart's Disease of Corn. *Phytopathology* **1935**, *25*, 371–372.
173. Okabe, N.; Goto, M. Bacteriophages of Plant Pathogens. *Annu. Rev. Phytopathol.* **1963**, *1*, 397–418. [CrossRef]
174. McCallin, S.; Brüssow, H. Phage Therapy: An Alternative or Adjunct to Antibiotics? *Emerg. Top. Life Sci.* **2017**, *1*, 105–116. [CrossRef]
175. Richards, G.P. Bacteriophage Remediation of Bacterial Pathogens in Aquaculture: A Review of the Technology. *Bacteriophage* **2014**, *4*, e975540. [CrossRef]
176. Gutiérrez, D.; Rodríguez-Rubio, L.; Fernández, L.; Martínez, B.; Rodríguez, A.; García, P. Applicability of Commercial Phage-Based Products against *Listeria monocytogenes* for Improvement of Food Safety in Spanish Dry-Cured Ham and Food Contact Surfaces. *Food Control* **2017**, *73*, 1474–1482. [CrossRef]
177. Jikia, D.; Chkhaidze, N.; Imedashvili, E.; Mgaloblishvili, I.; Tsitlanadze, G.; Katsarava, R.; Morris, J.G.; Sulakvelidze, A. The Use of a Novel Biodegradable Preparation Capable of the Sustained Release of Bacteriophages and Ciprofloxacin, in the Complex Treatment of Multidrug-Resistant *Staphylococcus aureus*—Infected Local Radiation Injuries Caused by Exposure to Sr90. *Clin. Exp. Dermatol.* **2005**, *30*, 23–26. [CrossRef]
178. Balogh, B.; Jones, J.; Iriarte, F.; Momol, M. Phage Therapy for Plant Disease Control. *Curr. Pharm. Biotechnol.* **2010**, *11*, 48–57. [CrossRef] [PubMed]
179. Jones, J.B.; Svircev, A.M.; Obradović, A.Ž. Crop Use of Bacteriophages. In *Bacteriophages*; Harper, D.R., Abedon, S.T., Burrowes, B.H., McConville, M.L., Eds.; Springer: Cham, Switzerland, 2021; pp. 1–18. [CrossRef]
180. Peterson, E.; Kaur, P. Antibiotic Resistance Mechanisms in Bacteria: Relationships between Resistance Determinants of Antibiotic Producers, Environmental Bacteria, and Clinical Pathogens. *Front. Microbiol.* **2018**, *9*, 2928. [CrossRef] [PubMed]
181. Schroth, M.N. Streptomycin Resistance in *Erwinia amylovora*. *Phytopathology* **1979**, *69*, 565–568. [CrossRef]
182. Stall, R.E.; Loschke, D.C.; Rice, R.W. Conjugational Transfer of Copper Resistance and Avirulence to Pepper within Strains of *Xanthomonas campestris* pv. *vesicatoria*. *Phytopathology* **1984**, *74*, 797.
183. The Commission of the European Communities. EUR-Lex-02004D0129-20050820-EN-EUR-Lex. 2005. Available online: https://eur-lex.europa.eu/legal-content/EN/TXT/?uri=CELEX%3A02004D0129-20050820 (accessed on 15 April 2023).
184. Di Lallo, G.; Evangelisti, M.; Mancuso, F.; Ferrante, P.; Marcelletti, S.; Tinari, A.; Superti, F.; Migliore, L.; D'Addabbo, P.; Frezza, D.; et al. Isolation and Partial Characterization of Bacteriophages Infecting *Pseudomonas syringae* pv. *actinidiae*, Causal Agent of Kiwifruit Bacterial Canker. *J. Basic Microbiol.* **2014**, *54*, 1210–1221. [CrossRef]
185. van Charante, F.; Holtappels, D.; Blasdel, B.; Burrowes, B. Isolation of Bacteriophages. In *Bacteriophages*; Springer International: Cham, Switzerland, 2019; pp. 1–32.
186. Loc-Carrillo, C.; Abedon, S.T. Pros and Cons of Phage Therapy. *Bacteriophage* **2011**, *1*, 111–114. [CrossRef]
187. Obradovic, A.; Jones, J.B.; Momol, M.T.; Balogh, B.; Olson, S.M. Management of Tomato Bacterial Spot in the Field by Foliar Applications of Bacteriophages and SAR Inducers. *Plant Dis.* **2004**, *88*, 736–740. [CrossRef]
188. Obradovic, A.; Jones, J.B.; Momol, M.T.; Olson, S.M.; Jackson, L.E.; Balogh, B.; Guven, K.; Iriarte, F.B. Integration of Biological Control Agents and Systemic Acquired Resistance Inducers Against Bacterial Spot on Tomato. *Plant Dis.* **2005**, *89*, 712–716. [CrossRef] [PubMed]
189. Pires, D.P.; Oliveira, H.; Melo, L.D.R.; Sillankorva, S.; Azeredo, J. Bacteriophage-Encoded Depolymerases: Their Diversity and Biotechnological Applications. *Appl. Microbiol. Biotechnol.* **2016**, *100*, 2141–2151. [CrossRef]
190. Milho, C.; Silva, M.D.; Sillankorva, S.; Harper, D.R. Biofilm Applications of Bacteriophages. In *Bacteriophages*; Springer International: Cham, Switzerland, 2019; pp. 1–35.
191. Ni, P.; Wang, L.; Deng, B.; Jiu, S.; Ma, C.; Zhang, C.; Almeida, A.; Wang, D.; Xu, W.; Wang, S. Combined Application of Bacteriophages and Carvacrol in the Control of *Pseudomonas syringae* pv. *actinidiae* Planktonic and Biofilm Forms. *Microorganisms* **2020**, *8*, 837. [CrossRef] [PubMed]
192. Vidaver, A.K.; Koski, R.K.; Van Etten, J.L. Bacteriophage Phi6: A Lipid-Containing Virus of *Pseudomonas phaseolicola*. *J. Virol.* **1973**, *11*, 799–805. [CrossRef] [PubMed]
193. Persley, Y.G.J.; Crosse, J.E. *A* Bacteriophage Specific to Race 2 of the Cherry Strain of *Pseudomonas morsprunorum*. *Ann. Appl. Biol.* **1978**, *89*, 219–222. [CrossRef]
194. Nordeen, R.O.; Morgan, M.K.; Currier, T.C. Isolation and Partial Characterization of Bacteriophages of the Phytopathogen *Pseudomonas syringae*. *Appl. Environ. Microbiol.* **1983**, *45*, 1890–1898. [CrossRef]
195. Minor, S.M.; Nordeen, R.O.; Paschall, C.E.; Minor, S.M.; Nordeen, R.O. Partial Characterization of Bacteriophages of *Pseudomonas syringae* pv. *Tomato*. *J. Ark. Acad. Sci.* **1996**, *50*, 28.
196. Yin, Y.; Ni, P.; Deng, B.; Wang, S.; Xu, W.; Wang, D. Isolation and Characterization of Phages *against Pseudomonas syringae* pv. *actinidiae*. *Acta Agric. Scand. B. Soil Plant Sci.* **2019**, *69*, 199–208. [CrossRef]

197. Yu, J.G.; Lim, J.A.; Song, Y.R.; Heu, S.; Kim, G.H.; Koh, Y.J.; Oh, C.S. Isolation and Characterization of Bacteriophages against *Pseudomonas syringae* pv. *actinidiae* Causing Bacterial Canker Disease in Kiwifruit. *J. Microbiol. Biotechnol.* **2015**, *26*, 385–393. [CrossRef] [PubMed]
198. Park, J.; Lim, J.-A.; Yu, J.-G.; Oh, C.-S. Genomic Features and Lytic Activity of the Bacteriophage PPPL-1 Effective against *Pseudomonas syringae* pv. *actinidiae*, a Cause of Bacterial Canker in Kiwifruit. *J. Microbiol. Biotechnol.* **2018**, *28*, 1542–1546. [CrossRef] [PubMed]
199. Pinheiro, L.A.M.; Pereira, C.; Barreal, M.E.; Gallego, P.P.; Balcão, V.M.; Almeida, A. Use of Phage Φ6 to Inactivate *Pseudomonas syringae* pv. *actinidiae* in Kiwifruit Plants: In Vitro and Ex Vivo Experiments. *Appl. Microbiol. Biotechnol.* **2020**, *104*, 1319–1330. [CrossRef] [PubMed]
200. Fiorillo, A.; Frezza, D.; Di Lallo, G.; Visconti, S. A Phage Therapy Model for the Prevention of *Pseudomonas syringae* pv. *actinidiae* Infection of Kiwifruit Plants. *Plant Dis.* **2023**, *107*, 267–271. [CrossRef] [PubMed]
201. Oueslati, M.; Holtappels, D.; Fortuna, K.; Hajlaoui, M.R.; Lavigne, R.; Sadfi-Zouaoui, N.; Wagemans, J. Biological and Molecular Characterization of the Lytic Bacteriophage SoKa against *Pseudomonas syringae* pv. *syringae*, Causal Agent of Citrus Blast and Black Pit in Tunisia. *Viruses* **2022**, *14*, 1949. [CrossRef]
202. Zhang, H.; Wu, H.; Xia, H.; Zhong, C.; Li, L.; Zeng, C. Genomic Characterization of Two Nickie-like Bacteriophages That Infect the Kiwifruit Canker Phytopathogen *Pseudomonas syringae* pv. *actinidiae*. *Arch. Virol.* **2022**, *167*, 1713–1715. [CrossRef] [PubMed]
203. Korniienko, N.; Kharina, A.; Zrelovs, N.; Jindřichová, B.; Moravec, T.; Budzanivska, I.; Burketová, L.; Kalachova, T. Isolation and Characterization of Two Lytic Phages Efficient Against Phytopathogenic Bacteria from *Pseudomonas* and *Xanthomonas* Genera. *Front. Microbiol.* **2022**, *13*, 853593. [CrossRef] [PubMed]
204. Bai, J.; Liu, Y.; Liu, M.; Luo, S.; Cheng, Y.; Li, G.; Liu, C.; Wen, S.; Xia, M.; He, X.; et al. Application of Phage Therapy against Red-Fleshed Kiwifruit Canker. *Biol. Control* **2022**, *169*, 104893. [CrossRef]
205. Kazantseva, O.A.; Buzikov, R.M.; Pilipchuk, T.A.; Valentovich, L.N.; Kazantsev, A.N.; Kalamiyets, E.I.; Shadrin, A.M. The Bacteriophage Pf-10—A Component of the Biopesticide "Multiphage" Used to Control Agricultural Crop Diseases Caused by *Pseudomonas syringae*. *Viruses* **2022**, *14*, 42. [CrossRef] [PubMed]
206. Liu, Y.; Liu, M.; Hu, R.; Bai, J.; He, X.; Jin, Y. Isolation of the Novel Phage PHB09 and Its Potential Use against the Plant Pathogen *Pseudomonas syringae* pv. *actinidiae*. *Viruses* **2021**, *13*, 2275. [CrossRef]
207. Martino, G.; Holtappels, D.; Vallino, M.; Chiapello, M.; Turina, M.; Lavigne, R.; Wagemans, J.; Ciuffo, M. Molecular Characterization and Taxonomic Assignment of Three Phage Isolates from a Collection Infecting *Pseudomonas syringae* pv. *actinidiae* and *P. syringae* pv. *phaseolicola* from Northern Italy. *Viruses* **2021**, *13*, 2083. [CrossRef]
208. Akbaba, M.; Ozaktan, H. Evaluation of Bacteriophages in the Biocontrol of *Pseudomonas syringae* pv. *syringae* Isolated from Cankers on Sweet Cherry (*Prunus avium* L.) in Turkey. *Egypt. J. Biol. Pest Control* **2021**, *31*, 35. [CrossRef]
209. Holtappels, D.; Kerremans, A.; Busschots, Y.; Vaerenbergh Van, J.V.; Maes, M.; Lavigne, R.; Wagemans, J. Preparing for the KIL: Receptor Analysis of *Pseudomonas syringae* pv. *porri* Phages and Their Impact on Bacterial Virulence. *Int. J. Mol. Sci.* **2020**, *21*, 2930. [CrossRef]
210. Jørgensen, J.B.; Djurhuus, A.M.; Carstens, A.B.; Kot, W.; Neve, H.; Morris, C.E.; Hansen, L.H. Presentation of Three Novel Tailed Phages Targeting Multiple Strains of *Pseudomonas syringae*. *Phage* **2020**, *1*, 245–250. [CrossRef] [PubMed]
211. Yu, P.; Mathieu, J.; Li, M.; Dai, Z.; Alvarez, P.J.J. Isolation of Polyvalent Bacteriophages by Sequential Multiple-Host Approaches. *Appl. Environ. Microbiol.* **2015**, *82*, 808–815. [CrossRef] [PubMed]
212. Thomas, M.D. Bacteriophages from Sewage Specific for Fluorescent Phytopathogenic Pseudomonads. *Phytopathology* **1983**, *73*, 403. [CrossRef]
213. Brockhurst, M.A.; Koskella, B.; Zhang, Q.-G. Bacteria-Phage Antagonistic Coevolution and the Implications for Phage Therapy. In *Bacteriophages*; Springer International: Cham, Switzerland, 2017; pp. 1–21. [CrossRef]
214. Zhang, H.; Yang, Z.; Zhou, Y.; Bao, H.; Wang, R.; Li, T.; Pang, M.; Sun, L.; Zhou, X. Application of a Phage in Decontaminating *Vibrio parahaemolyticus* in Oysters. *Int. J. Food Microbiol.* **2018**, *275*, 24–31. [CrossRef]
215. Tom, E.F.; Molineux, I.J.; Paff, M.L.; Bull, J.J. Experimental Evolution of UV Resistance in a Phage. *PeerJ* **2018**, *6*, e5190. [CrossRef]
216. Iriarte, F.B.; Obradović, A.; Wernsing, M.H.; Jackson, L.E.; Balogh, B.; Hong, J.A.; Momol, M.T.; Jones, J.B.; Vallad, G.E. Soil-Based Systemic Delivery and Phyllosphere in vivo Propagation of Bacteriophages. *Bacteriophage* **2012**, *2*, e23530. [CrossRef]
217. Kolozsváriné Nagy, J.; Schwarczinger, I.; Künstler, A.; Pogány, M.; Király, L. Penetration and Translocation of *Erwinia amylovora* Specific Bacteriophages in Apple—A Possibility of Enhanced Control of Fire Blight. *Eur. J. Plant Pathol.* **2015**, *142*, 815–827. [CrossRef]
218. Rao, P.S.; Srivastava, H.C. *Others Industrial Gums*; Whistler, R.L., Ed.; Academic Press: Cambridge, MA, USA, 1973; pp. 372–411.
219. Jackson, L.R.E. Bacteriophage Prevention and Control of Harmful Plant Bacteria. U.S. Patent US4828999A, 9 May 1989.
220. Holtappels, D.; Fortuna, K.; Lavigne, R.; Wagemans, J. The Future of Phage Biocontrol in Integrated Plant Protection for Sustainable Crop Production This Review Comes from a Themed Issue on Nanobiotechnology-Phage Therapy. *Curr. Opin. Biotechnol.* **2021**, *68*, 60–71. [CrossRef]
221. Jończyk-Matysiak, E.; Łodej, N.; Kula, D.; Owczarek, B.; Orwat, F.; Międzybrodzki, R.; Neuberg, J.; Bagińska, N.; Weber-Dąbrowska, B.; Górski, A. Factors Determining Phage Stability/Activity: Challenges in Practical Phage Application. *Expert. Rev. Anti Infect. Ther.* **2019**, *17*, 583–606. [CrossRef]

222. Lin, K.; Schulte, C.R.; Marr, L.C. Survival of MS2 and Φ6 Viruses in Droplets as a Function of Relative Humidity, PH, and Salt, Protein, and Surfactant Concentrations. *PLoS ONE* **2020**, *15*, e0243505. [CrossRef] [PubMed]
223. Sieiro, C.; Areal-hermida, L.; Pichardo-gallardo, Á.; Almuiña-González, R.; de Miguel, T.; Sánchez, S.; Sánchez-pérez, Á.; Villa, T.G. A Hundred Years of Bacteriophages: Can Phages Replace Antibiotics in Agriculture and Aquaculture? *Antibiotics* **2020**, *9*, 493. [CrossRef] [PubMed]