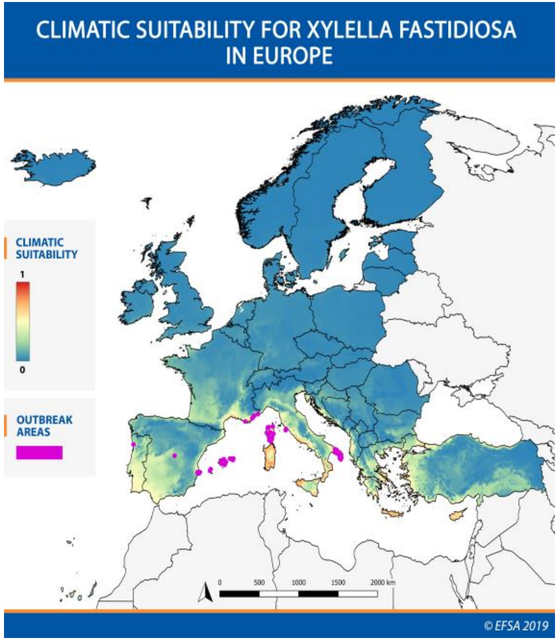
3rd European Conference on
Xylella fastidiosa and XF-ACTORS final meeting

Isolation and characterization of bacteriophages against *Xylella fastidiosa*

M.L. Domingo-Calap, C.M. Aure, I. Navarro-Herrero, P. Domingo-Calap, E. Marco-Noales

INTRODUCTION

Importance of *Xylella fastidiosa*



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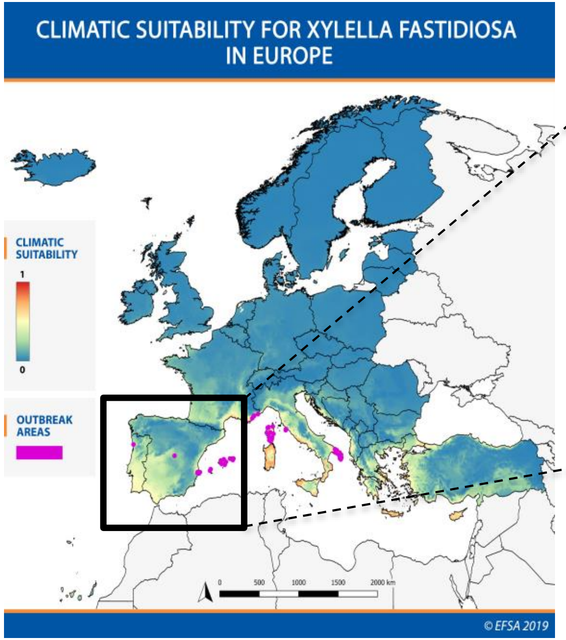

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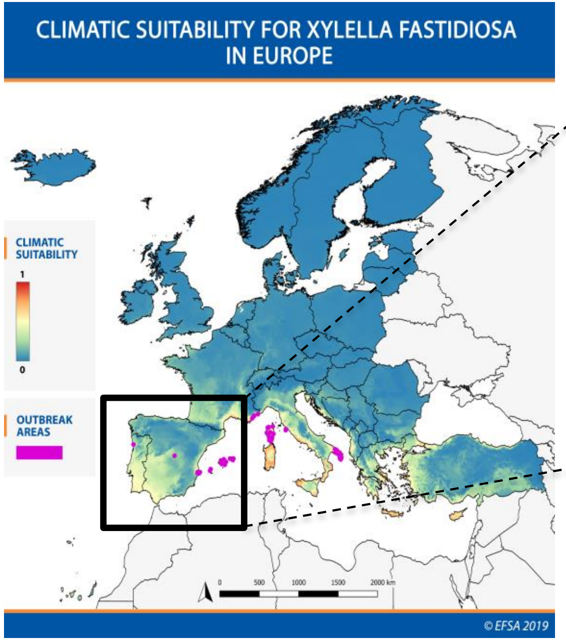


Control measures

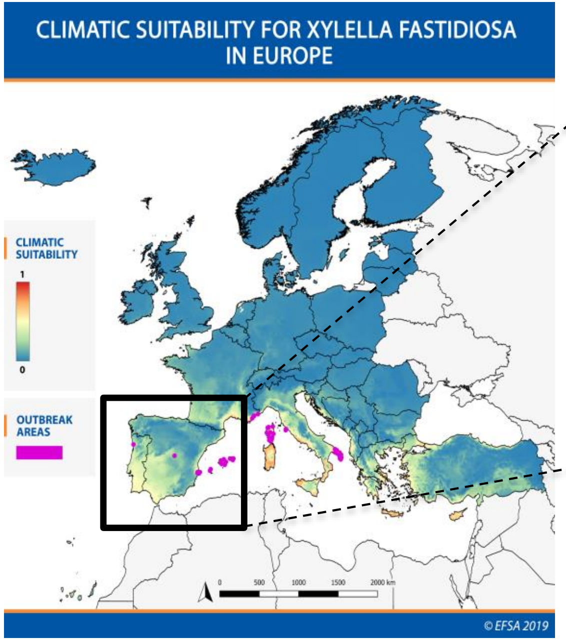
- Chemical control

- Erradication of infected plants

➡ Develop new control strategies

INTRODUCTION

Importance of *Xylella fastidiosa*



Control measures

- Chemical control

- Erradication of infected plants

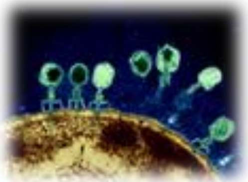
➡ Develop new control strategies

To search specific bacteriophages for *X. fastidiosa*

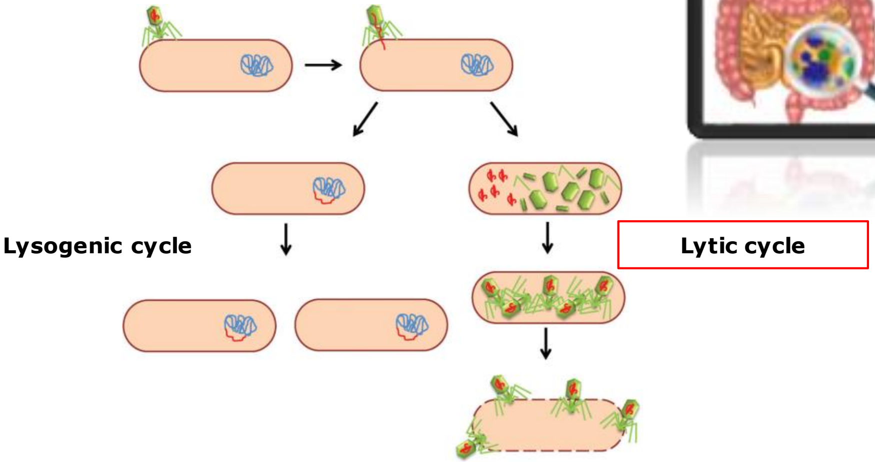
INTRODUCTION

PHAGES



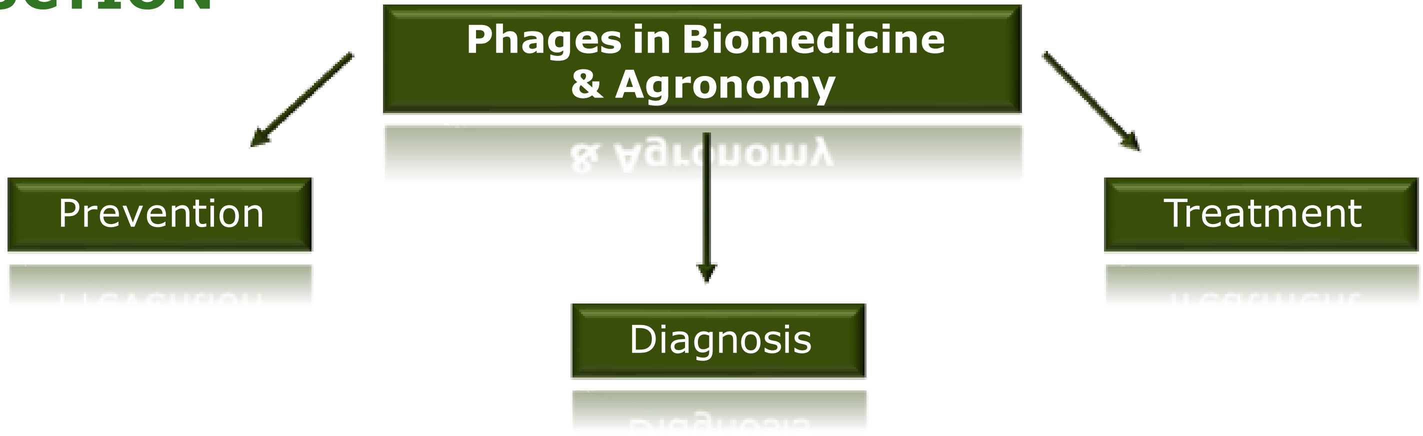



- ➢ Bacteriophages: natural predators of bacteria
- ➢ Are the most abundant biological entity in the biosphere with an estimated number of 10^{31}
  - ➢ 10^{7} phages/mL in aquatic environment and 10^{9} phages in sediments
  - ➢ Up to ~ 10^{10} per gram of soil
  - ➢ Human gut: 10^{15} particles, >100 species
- ➢ At least 10 times more phages than bacteria
- ➢ Highly variable
- ➢ Biological cycle: lytic vs. lysogenic



Domingo-Calap et al. HLA 2016

INTRODUCTION



INTRODUCTION

Phages in Biomedicine & Agronomy

- Prevention
- Diagnosis
- Treatment

Comparison of some properties of drug-antibiotic treatments and phage therapy against bacterial infections

	Drugs – antibiotics	Phages
Ecological safety	No	Yes
Side effects	Yes	No (thus far)
Specificity	Low – broad range	High – limited range
Induction of resistance	High	Low
Cost	Usually high	Low
Industrial production	Long	Fast
Patentable	Yes	No
Research in the field	Many publications Many clinical trials	Lack of controlled studies

Domingo-Calap et al. HLA 2016

INTRODUCTION

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Domingo-Calap et al. HLA 2016


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Health | Coronavirus

Phage therapy: 'Viral cocktail saved my daughter's life'

By James Gallagher
Health and science correspondent, BBC News

8 May 2019

JO CARNELL HOLDAWAY

Isabelle Carnell-Holdaway is now 17 and learning to drive

An experimental cocktail of viruses has saved the life of a teenager who had a deadly and seemingly untreatable infection.

Isabelle's body was being attacked by bacteria and she was given less than a 1% chance of survival.

But doctors at Great Ormond Street Hospital attempted an untested "phage therapy", which uses viruses to infect and kill bacteria.

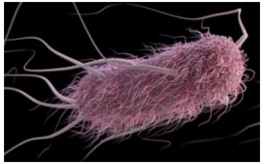
Isabelle is now learning to drive and studying for her A-levels.

Experts said the case was "enormously exciting" and showed the potential for treating other dangerous infections with phage.

INTRODUCTION

Examples of phage therapy in EU

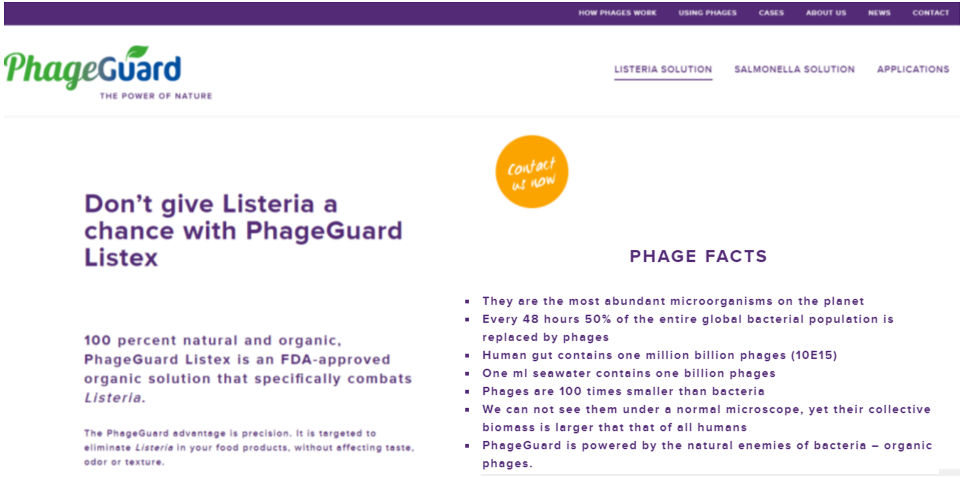
Phagoburn: *E. coli* and *P. aeruginosa*

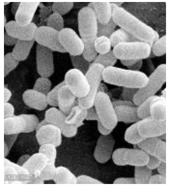


E.coli

P. aeruginosa

Listex: *Listeria sp.*





Listeria sp.

INTRODUCTION Examples of bacteriophage biocontrol experiments

Pathogen	Host	Disease	Information	Reference
Pectobacterium carotovorum ssp. carotovorum, *Pectobacterium wasabiae*, *Dickeya solani*	Potato	Soft rot	Bioassays with phage ΦPD10.3 and ΦPD23.1 could reduce severity of soft rot of tubers by 80% on potato slices and 95% with whole tubers from a mixed pathogen infection.	Czajkowski et al., 2015
Dickeya solani	Potato	Soft rot/Blackleg	Phage vB_DsoM_LIMEstone1 and vB_DsoM_LIMEstone2 reduced soft rot of inoculated tubers in bioassays and in field trials which produced a potato crop with higher yields.	Adriaenssens et al., 2012
Dickeya solani	Potato	Soft rot	Bioassays with phage ΦD1, ΦD2, ΦD3, ΦD4, ΦD5, ΦD7, ΦD9, ΦD10, ΦD11 could reduce incidence of soft rot by up to 30–70% on co-inoculated potato slices with pathogen and phage.	Czajkowski et al., 2014
Streptomyces scabies	Potato	Common scab	Seed tubers treated with phage ΦAS1 resulted in producing tuber progeny with reduced levels of surface lesion of scab (1.2%) compared with tubers harvested from non -treated seed tubers (23%).	McKenna et al., 2001
Ralstonia solanacearum	Tomato	Bacterial wilt	Tomato plants treated with phage ΦRSL1 showed no symptoms of bacterial wilt during the experimental period; whereas all untreated plants showed wilting 18 days post infection.	Fujiwara et al., 2011
Ralstonia solanacearum	Tomato	Bacteria wilt	Simultaneous treatment of phage PE204 with *R. solanacearum* of the rhizosphere of tomato completely inhibited bacterial wilt. However, pre-treatment with phage before the inoculation of pathogen was not effective with control of bacterial wilt, whereas post treatment of PE204 delayed disease development.	Bae et al., 2012
Xanthomonas campestris pv. vesicatoria	Tomato	Bacterial spot	Greenhouse experiments with formulated phage cocktails could reduce disease severity with formulated phage cocktails providing better protection in comparison to unformulated. A similar effect was found in three consecutive field trials.	Balogh et al., 2003
Xanthomonas campestris pv. vesicatoria	Tomato	Bacterial spot	In field experiments phage treatment was comparable to disease control with copper-mancozeb. Combination of phage and plant activator (ASM) resulted in enhanced control.	Obradovic et al., 2004
Xylella fastidiosa	Grapevines	Pierce's Disease	*X. fastidiosa* levels in grapevines were significantly reduced on pre and post inoculation of a four phage (*Sano, Salvo, Prado and Paz*) cocktail. Pierce disease symptoms could be stopped using phage treatment post infection as well as applying phage prophylactically to grapevines.	Das et al., 2015
Xanthomonas axonopodis pv. *allii*	Onion	*Xanthomonas* leaf blight of onion	Field trial showed that weekly and biweekly applications of phage could reduce disease severity, a result which was comparable to treatments of weekly applications of copper-mancozeb.	Lang et al., 2007
Pectobacterium carotovorum ssp. *carotovorum*	Lettuce	Soft rot	Green house trials showed that phage PP1 could significantly reduce disease development on lettuce plants.	Lim et al., 2013

Buttimer *et al.*, 2017. doi: *10.3389/fmicb.2017.00034*

OBJECTIVE

To isolate specific phages against *X. fastidiosa*

XF ACTORS

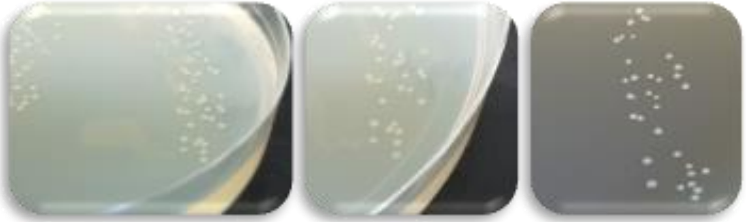
Xylella Fastidiosa Active Containment Through a multidisciplinary-Oriented Research Strategy

WP 6-TASK 6.2 (IVIA)

MATERIAL & METHODS

Double layer assays

- Specific culture medium
- Slow growth



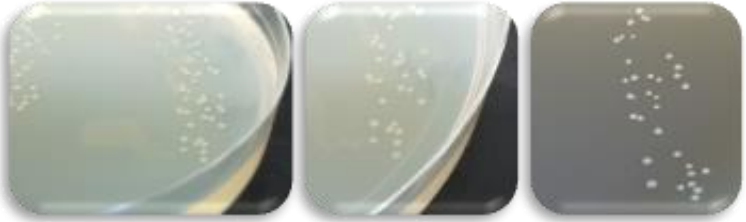
MATERIAL & METHODS

Double layer assays

- Specific culture medium
- Slow growth

⇩

<u>Surrogate host</u>



***Xanthomonas* spp**

- ***X. arboricola* pv. *juglandis* (IVIA 1317-1a)** — **XAJ**
- ***X. axonopodis* pv. *phaseoli* (CECT 914)** — **XAP**

Source: IVIA — *X. arboricola* pv. *juglandis*

Source: EPPO — *X. axonopodis* pv. *phaseoli*

MATERIAL & METHODS

SAMPLES

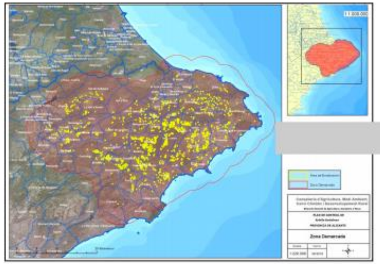
Vegetal samples infected by *Xanthomonas* spp.

Plant samples infected by *Xylella fastidiosa* from Balearic islands and Alicante outbreaks

Soil samples from Balearic islands and Alicante outbreaks

River water from Balearic islands and Alicante outbreaks

Alicante Outbreak (Spain)

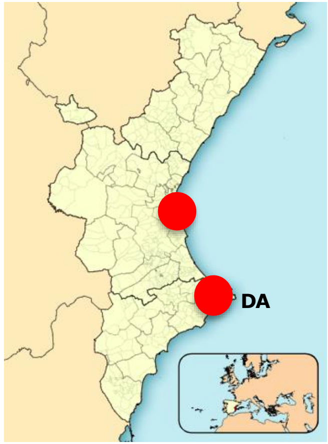


Number of samples	Plant species	Origin	Infected by
10	*Prunus dulcis*	Alicante, Comunidad Valenciana (Spain)	*Xylella fastidiosa*
5	Soil	Alicante, Comunidad Valenciana (Spain)	Area with almond trees infected by *X. fastidiosa*
5	Water from irrigation channel	Alicante, Comunidad Valenciana (Spain)	Area with almond trees infected by *X. fastidiosa*

Number of samples	Plant species	Origin	Infected by
3	*Juglans* sp.	Comunidad Valenciana (Spain)	*Xanthomonas arboricola* pv. *juglandis*
2	*Acacia saligna*		
1	*Lavandula dentata*		
3	*Nerium oleander*		
18	*Olea europaea* var. *sylvestris*		
9	*Olea europaea* var. *europaea*	Balearic Islands (Spain)	*Xylella fastidiosa*
11	*Polygala myrtifolia*		
3	*Prunus avium*		
1	*Prunus domestica*		
6	*Prunus dulcis*		
2	*Rosmarinus officinalis*		

MATERIAL & METHODS

SAMPLES
Vegetal samples infected by *Xanthomonas* spp.
Plant samples infected by *Xylella fastidiosa* from Balearic islands and Alicante outbreaks
Soil samples from Balearic islands and Alicante outbreaks
River water from Balearic islands and Alicante outbreaks
Wastewater samples from Valencia



WASTEWATER SAMPLES

α49: Pinedo: entrada 22-09-2020
α50: Pinedo: muestra puntual 24-09-2020
α51: Quart-Benáger: entrada 22-09-2020
α52: Quart-Benáger: muestra puntual 23-09-2020

Dr. Pilar Domingo-Calap
Enviromental and biomedical viruses **(EnBiVirLab)**
I2SYSBIO (UV-CSIC)

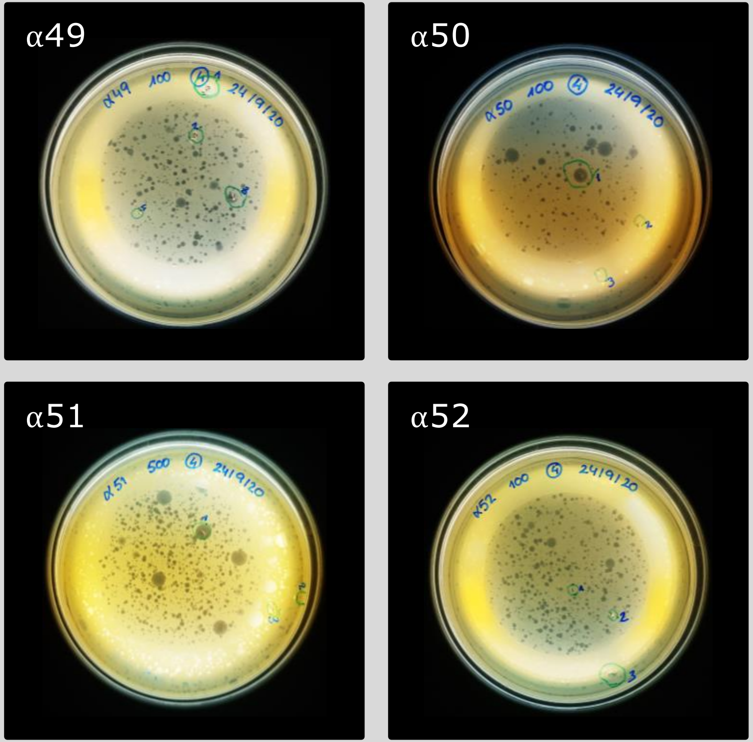
RESULTS

XAJ

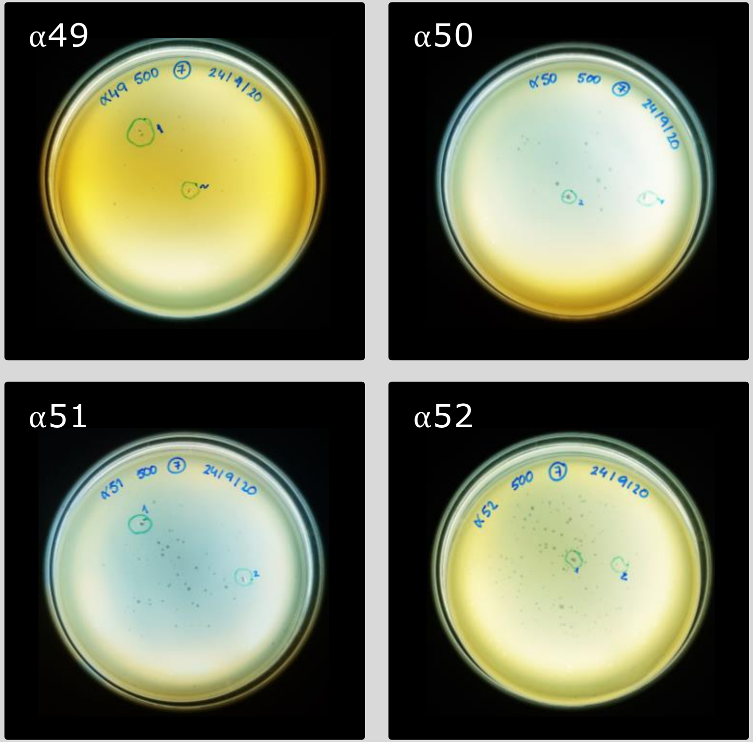
Bacteriophages present in wastewater samples

XAP

***X. arboricola* pv. *juglandis* (IVIA 1317-1a)**



***X. axonopodis* pv. *phaseoli* (CECT 914)**



RESULTS

Bacteriophages isolation

XAJ

X. arboricola pv. *juglandis* (IVIA 1317-1a)

PHAGE	SAMPLE	PLAQUE	DATE
P1	α49	C1	25/09/2020
P2	α49	C2	25/09/2020
P3	α49	C3	25/09/2020
P4	α49	C4	25/09/2020
P5	α50	C1	25/09/2020
P6	α50	C2	25/09/2020
P7	α50	C3	25/09/2020
P8	α51	C1	25/09/2020
P9	α51	C2	25/09/2020
P10-1	α51	C3*	25/09/2020
P10-2	α51	C3*	25/09/2020
P11	α52	C1	25/09/2020
P12	α52	C2	25/09/2020
P13	α52	C3	25/09/2020

XAP

X. axonopodis pv. *phaseoli* (CECT 914)

PHAGE	SAMPLE	PLAQUE	DATE
P14	α49	C1	25/09/2020
P15	α49	C2	25/09/2020
P16	α50	C1	25/09/2020
P17	α50	C2	25/09/2020
P18	α51	C1	25/09/2020
P19	α51	C2	25/09/2020
P20	α52	C1	25/09/2020
P21	α52	C2	25/09/2020

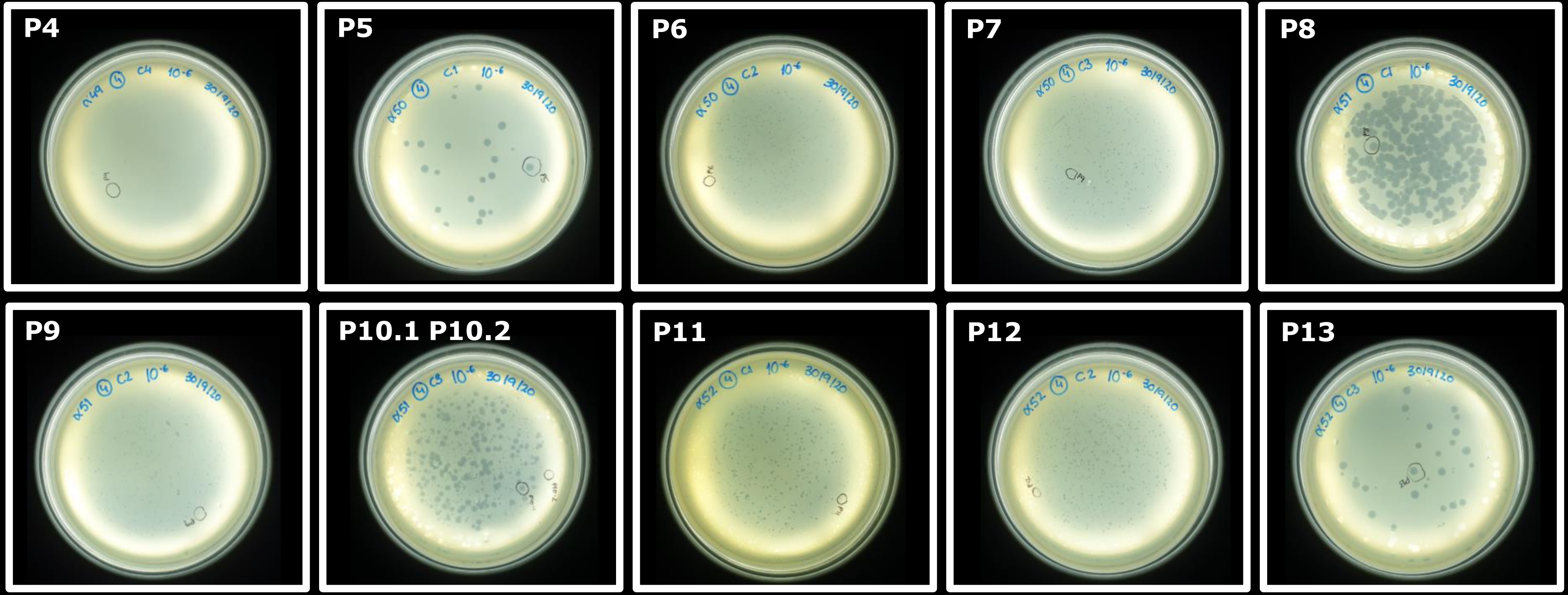
WASTEWATER SAMPLES

α49: Pinedo 1 entrada 22-09-2020
α50: Pinedo 1 muestra puntual 24-09-2020
α51: Quart-Benáger entrada 22-09-2020
α52: Quart-Benáger muestra puntual 23-09-2020

RESULTS

XAJ

PHAGES ISOLATED IN
X. arboricola* pv. *juglandis
(IVIA 1317-1a)

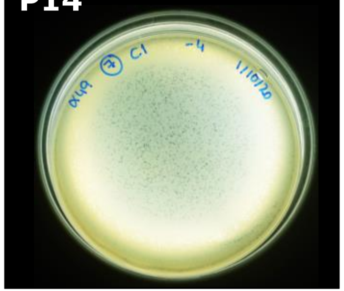


RESULTS

XAP

PHAGES ISOLATED IN
X. axonopodis* pv. *phaseoli
(CECT 914)

P14



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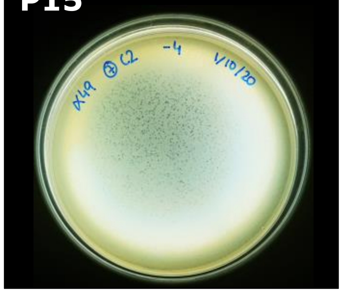

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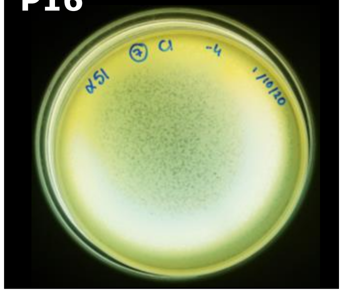

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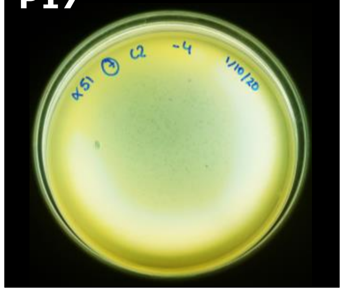

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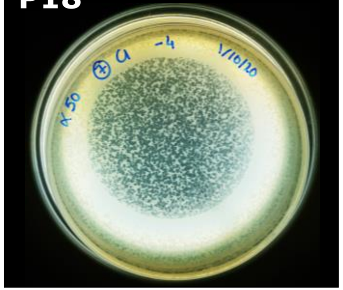

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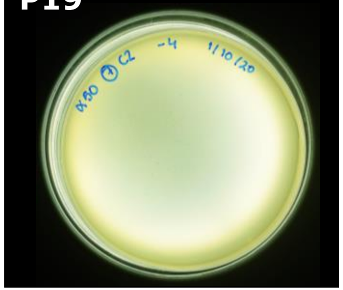

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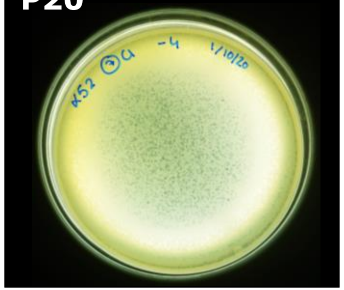
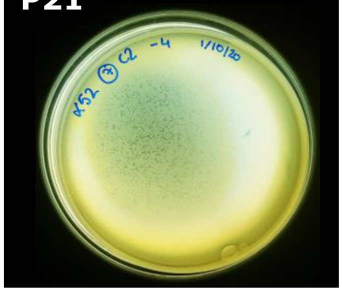


P21



RESULTS

Bacteriophage amplification

CONTROLS

500μl Bacterial OD_{600} 0,2

10μl bacteriophage

500μl Bacterial OD_{600} 0,2
+
10μl bacteriophage

24h at 25ºC

RESULTS

Bacteriophage amplification

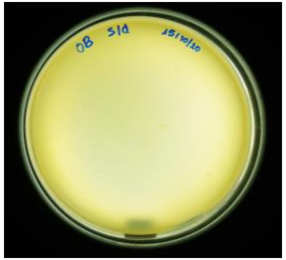
CONTROLS

500µl Bacterial OD_{600} 0,2

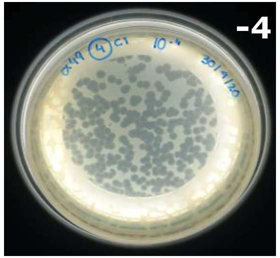
10µl bacteriophage

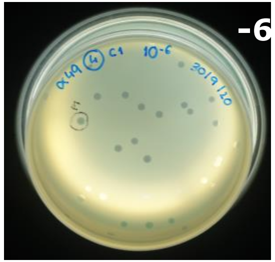
500µl Bacterial OD_{600} 0,2
+
10µl bacteriophage

24h at 25ºC

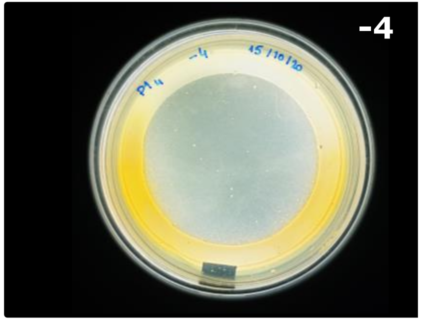


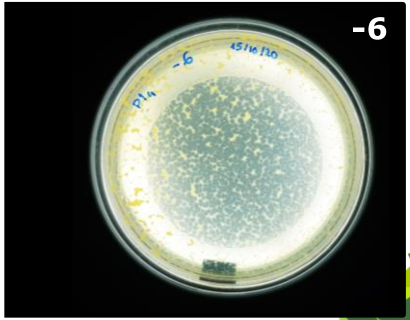
NOT AMPLIFIED PHAGE





AMPLIFIED PHAGE



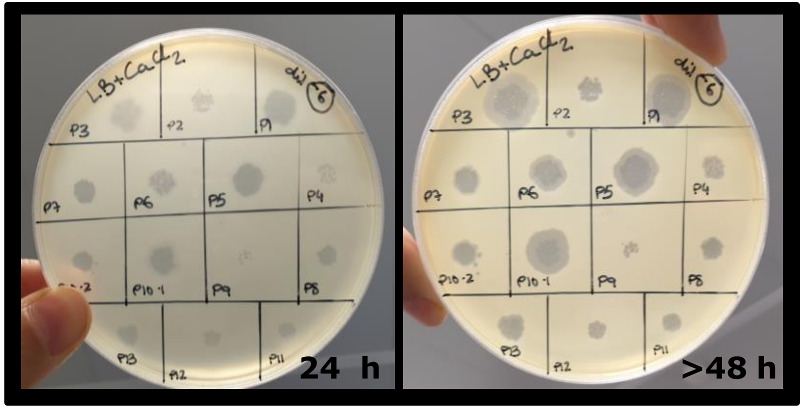


RESULTS

Amplified bacteriophages: Spot test

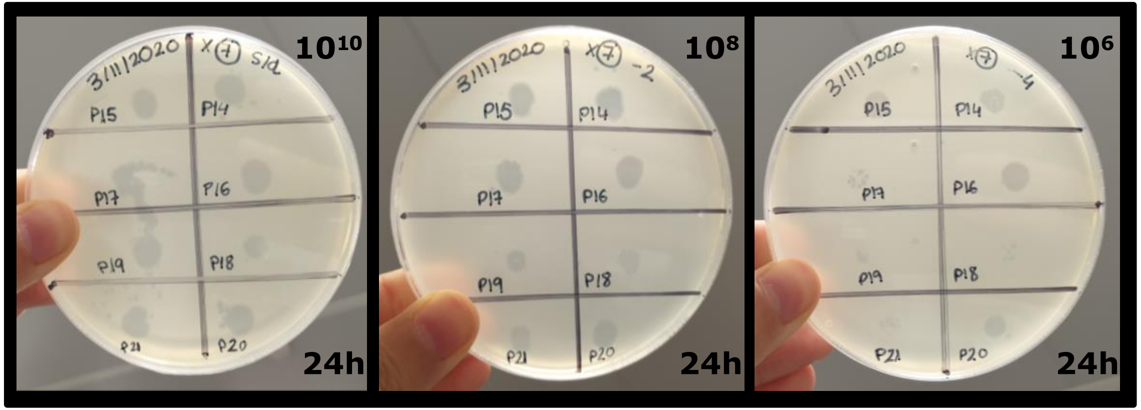
X. arboricola pv. *juglandis*
(IVIA 1317-1a)

XAJ



X. axonopodis pv. *phaseoli*
(CECT 914)

XAP



RESULTS

Host range

Xanthomonas spp.

SPECIES	STRAIN
Xanthomonas arboricola pv. juglandis	CFBP 2528
Xanthomonas arboricola pv. juglandis	IVIA 2499.1
Xanthomonas arboricola pv. juglandis	IVIA 3114.1
Xanthomonas arboricola pv. juglandis	IVIA 3493
Xanthomonas arboricola pv. juglandis	IVIA 3494
Xanthomonas arborícola pv. juglandis	IVIA 4254.1
Xanthomonas citri subsp. *citri*	XCC176
Xanthomonas alfafae pv. *citrumelo*	IRAN 11
Xanthomonas axonopodis pv. fuscans	IVIA 1518 DA
Xanthomonas hortorum carotae	IVIA 9489-1
Xanthomonas hortorum carotae	IVIA 9489-2
Xanthomonas arboricola pv. populi	CFBP 3123
Xanthomonas euvesicatoria	IVIA 4928
Xanthomonas campestris pv. campestris	IVIA 2734-1
Xanthomonas oryzae	XOO93
Xanthomonas arboricola pv. corylina	CFBP 1846
Xanthomonas vesicatoria	CECT 792
Xanthomonas arboricola pv. prunicola	IVIA 3287-1
Xanthomonas arboricola pv. prunicola	IVIA 3287-2
Xanthomonas arboricola pv. frgariae	CFBP 6771
Xanthomonas axonopodis pv. phaseoli	NCPPB 381
Xanthomonas axonopodis pv. phaseoli	NCPPB 3035
Xanthomonas axonopodis pv. phaseoli	NCPPB 3660

SPECIES	STRAIN
Xanthomonas arboricola pv. *pruni*	IVIA 2626.1
Xanthomonas arboricola pv. *pruni*	IVIA 2647.1.2
Xanthomonas arboricola pv. *pruni*	IVIA 2649.1
Xanthomonas arboricola pv. *pruni*	IVIA 2667
Xanthomonas arborícola pv.pruni	IVIA 2758.1
Xanthomonas arborícola pv.pruni	IVIA 2795
Xanthomonas arboricola pv. *pruni*	IVIA2826-1
Xanthomonas arboricola pv. *pruni*	IVIA2826-2
Xanthomonas arboricola pv. *pruni*	IVIA2826-7
Xanthomonas arboricola pv. *pruni*	IVIA2826-8
Xanthomonas arboricola pv. *pruni*	IVIA2826-9
Xanthomonas arboricola pv. *pruni*	IVIA2826-10
Xanthomonas arboricola pv. *pruni*	IVIA2832-4b
Xanthomonas arborícola pv. pruni	IVIA2832-30a
Xanthomonas arboricola pv. *pruni*	IVIA3767-3
Xanthomonas arboricola pv. *pruni*	IVIA 4113
Xanthomonas arboricola pv. *pruni*	IVIA 4165.15
Xanthomonas arboricola pv. *pruni*	DAR 33337
Xanthomonas arborícola pv. pruni	DAR 33420
Xanthomonas arboricola pv. *pruni*	DAR 56679
Xanthomonas arboricola pv. *pruni*	DAR 56680
Xanthomonas arboricola pv. *pruni*	DAR 61729
Xanthomonas arboricola pv. *pruni*	DAR 69849

Strains susceptible to viral infection

RESULTS

Host range

Xanthomonas spp.

SPECIES	STRAIN
Xanthomonas arboricola pv. juglandis	CFBP 2528
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Xanthomonas arboricola pv. juglandis	IVIA 3114.1
Xanthomonas arboricola pv. juglandis	IVIA 3493
Xanthomonas arboricola pv. juglandis	IVIA 3494
Xanthomonas arborícola pv. juglandis	IVIA 4254.1
Xanthomonas citri subsp. *citri*	XCC176
Xanthomonas alfafae pv. *citrumelo*	IRAN 11
Xanthomonas axonopodis pv. fuscans	IVIA 1518 DA
Xanthomonas hortorum carotae	IVIA 9489-1
Xanthomonas hortorum carotae	IVIA 9489-2
Xanthomonas arboricola pv. populi	CFBP 3123
Xanthomonas euvesicatoria	IVIA 4928
Xanthomonas campestris pv. campestris	IVIA 2734-1
Xanthomonas oryzae	XOO93
Xanthomonas arboricola pv. corylina	CFBP 1846
Xanthomonas vesicatoria	CECT 792
Xanthomonas arboricola pv. prunicola	IVIA 3287-1
Xanthomonas arboricola pv. prunicola	IVIA 3287-2
Xanthomonas arboricola pv. frgariae	CFBP 6771
Xanthomonas axonopodis pv. phaseoli	NCPPB 381
Xanthomonas axonopodis pv. phaseoli	NCPPB 3035
Xanthomonas axonopodis pv. phaseoli	NCPPB 3660

SPECIES	STRAIN
Xanthomonas arboricola pv. *pruni*	IVIA 2626.1
Xanthomonas arboricola pv. *pruni*	IVIA 2647.1.2
Xanthomonas arboricola pv. *pruni*	IVIA 2649.1
Xanthomonas arboricola pv. *pruni*	IVIA 2667
Xanthomonas arborícola pv.pruni	IVIA 2758.1
Xanthomonas arborícola pv.pruni	IVIA 2795
Xanthomonas arboricola pv. *pruni*	IVIA2826-1
Xanthomonas arboricola pv. *pruni*	IVIA2826-2
Xanthomonas arboricola pv. *pruni*	IVIA2826-7
Xanthomonas arboricola pv. *pruni*	IVIA2826-8
Xanthomonas arboricola pv. *pruni*	IVIA2826-9
Xanthomonas arboricola pv. *pruni*	IVIA2826-10
Xanthomonas arboricola pv. *pruni*	IVIA2832-4b
Xanthomonas arborícola pv. pruni	IVIA2832-30a
Xanthomonas arboricola pv. *pruni*	IVIA3767-3
Xanthomonas arboricola pv. *pruni*	IVIA 4113
Xanthomonas arboricola pv. *pruni*	IVIA 4165.15
Xanthomonas arboricola pv. *pruni*	DAR 33337
Xanthomonas arborícola pv. pruni	DAR 33420
Xanthomonas arboricola pv. *pruni*	DAR 56679
Xanthomonas arboricola pv. *pruni*	DAR 56680
Xanthomonas arboricola pv. *pruni*	DAR 61729
Xanthomonas arboricola pv. *pruni*	DAR 69849

Strains susceptible to viral infection

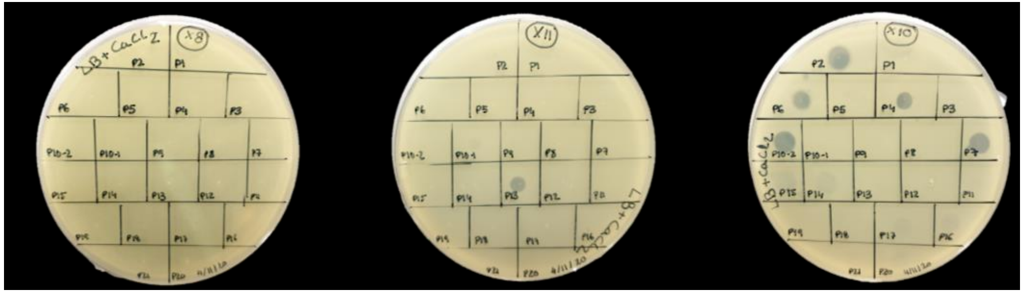
Other Phytopathogenic bacteria

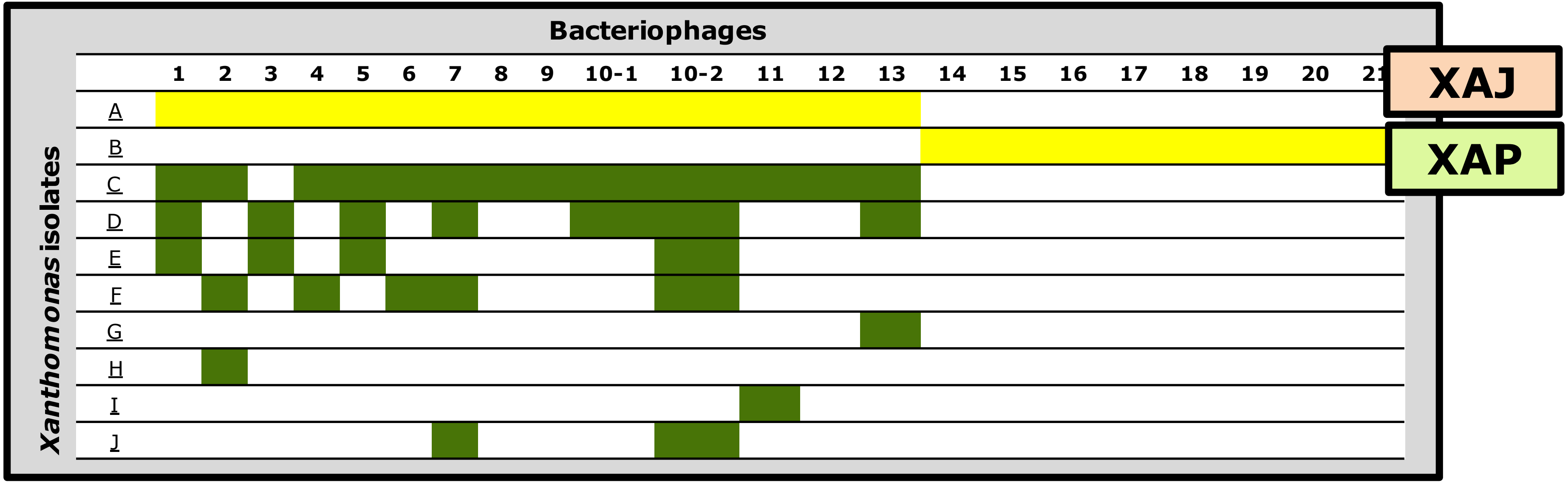
SPECIES	STRAIN
Agrobacterium tumefaciens	C-58
Agrobacterium rhizogenes	2649b
Agrobacterium vitis	IVIA 339-26
Dickeya chrysanthemi	IVIA 2048
Pseudomonas syringae pv. *syringae*	IVIA 773.1
Pseudomonas syringae pv. *syringae*	I015
Pseudomonas syringae pv. *syringae*	IVIA 4419.5
Pseudomonas savastanoi pv. *savastanoi*	IVIA 1628-3
Clavibacter michiganensis subsp. *michiganensis*	IVIA 2686-1

SPECIES	STRAIN
Clavivacter michiganensis subsp. *sepedoricus*	NCPPB 2140
Pseudomonas syringe actinidiae	IVIA 3918.6
Erwinia amylovora	IVIA 1892
Erwinia amylovora	CFBP1430
Lonsdalea quercina	IVIA 1618a
Pseudomonas syringe tomato	IVIA 1001.1a
Pectobacterium carotovorum	IVIA 3905.46
Xylophilus ampelinus	NCPPB 2217

RESULTS

Host range

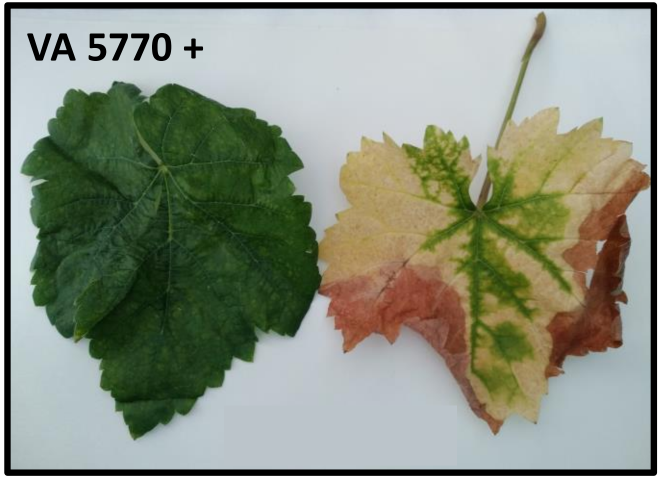


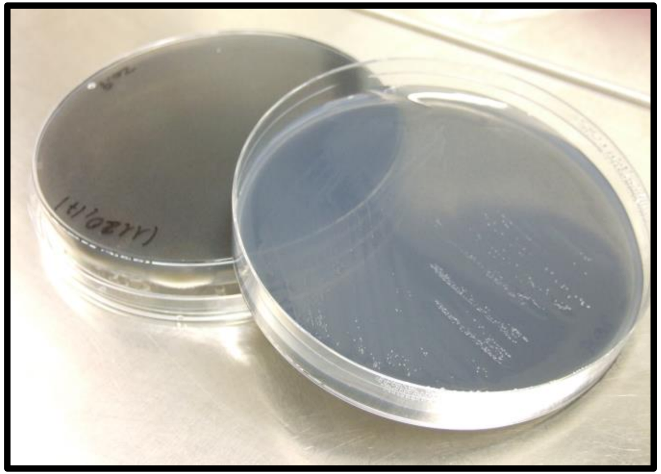


	Isolate	Specie
A	IVIA 1317-1a	*X. arboricola* pv. *juglandis*
B	CECT914	*X. axonopodis* pv. *phaseoli*
C	IVIA 4165.15	*X. arboricola* pv. *pruni*
D	IRAN11	*X. alfafae* pv. *citrumelo*
E	XCC 176	*X. citri* subsp. *citri*
F	IVIA 4928	*X. euvesicatoria*
G	IVIA 2734-1	*X. campestris* pv. *campestris*
H	XOO93	*X. oryzae*
I	CFBP6771	*X. arboricola* pv. *fragariae*
J	IVIA9489-1	*X. hortorum carotae*

RESULTS

Specie	subespecie	Strain	ST	Host	Origin	Year
Xylella fastidiosa	*multiplex*	IVIA 5901	ST 6	Almond	Alicante (Spain)	2017
	fastidiosa	IVIA 5770	ST 1	Grapevine	Mallorca (Spain)	2017



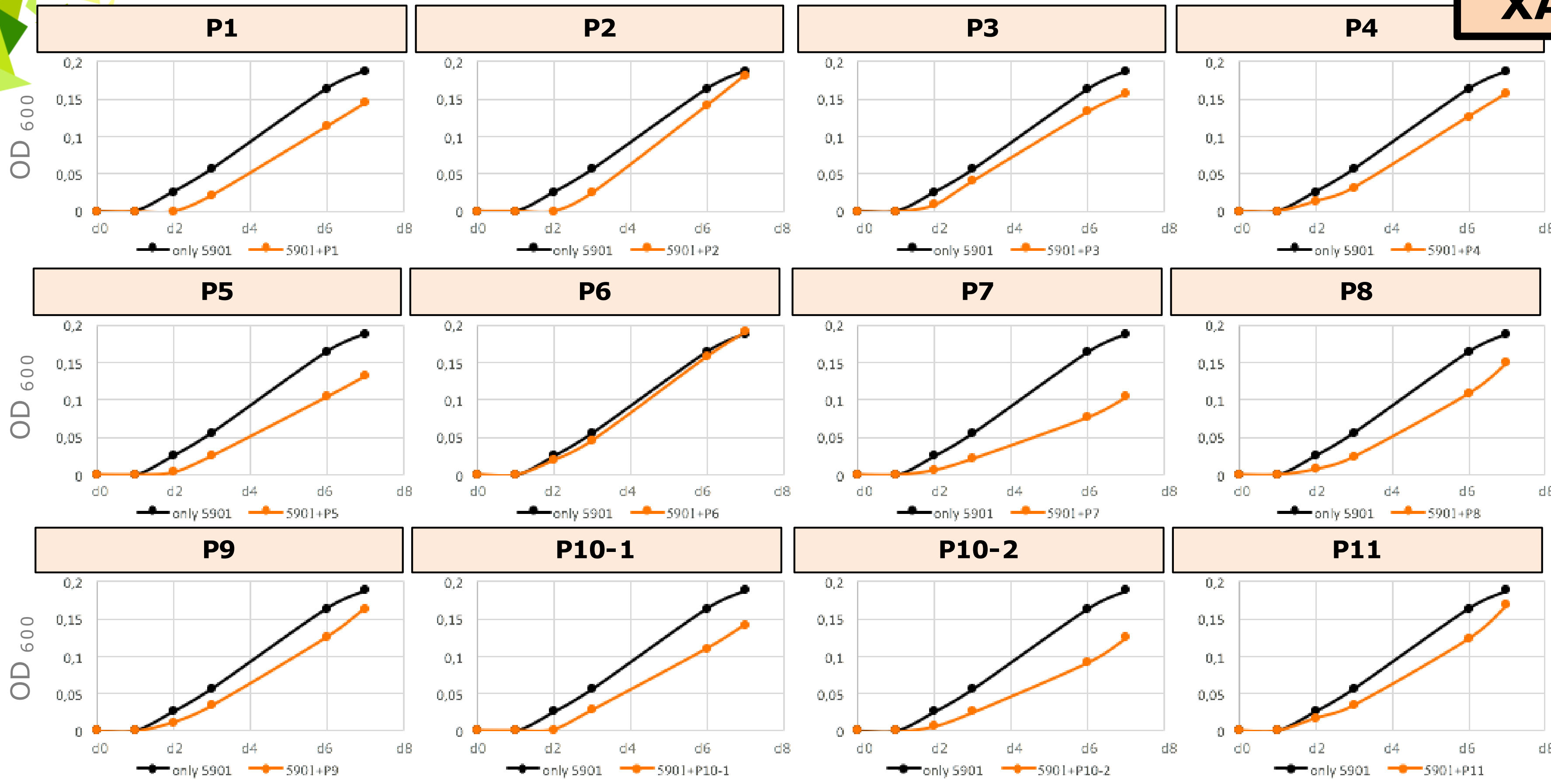


RESULTS

X. fastidiosa subsp. *multiplex* **IVIA 5901**

Host range

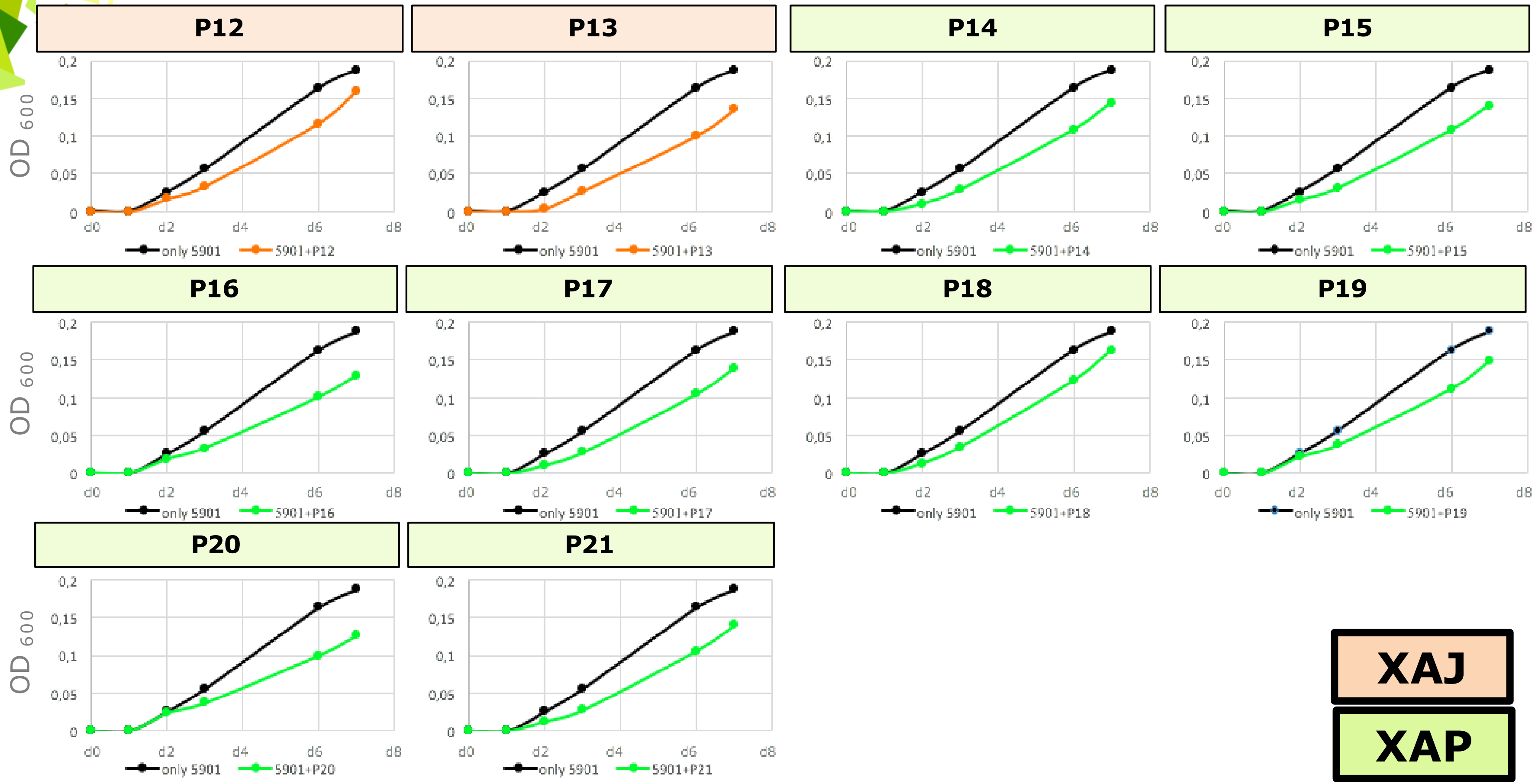
XAJ



RESULTS

X. fastidiosa subsp. *multiplex* **IVIA 5901**

Host range



XAJ

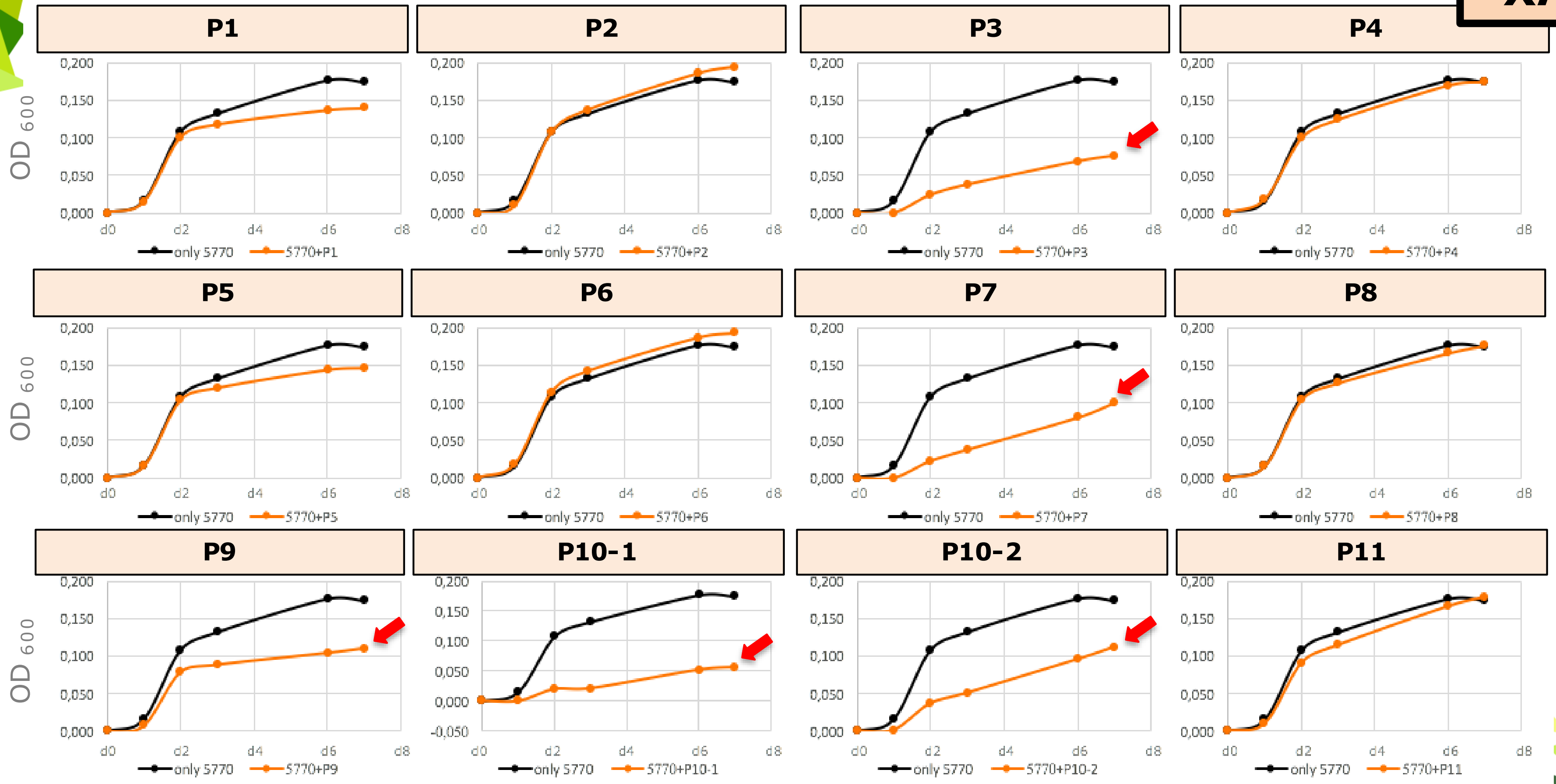
XAP

RESULTS

X. fastidiosa subsp. *fastidiosa* **IVIA 5770**

Host range

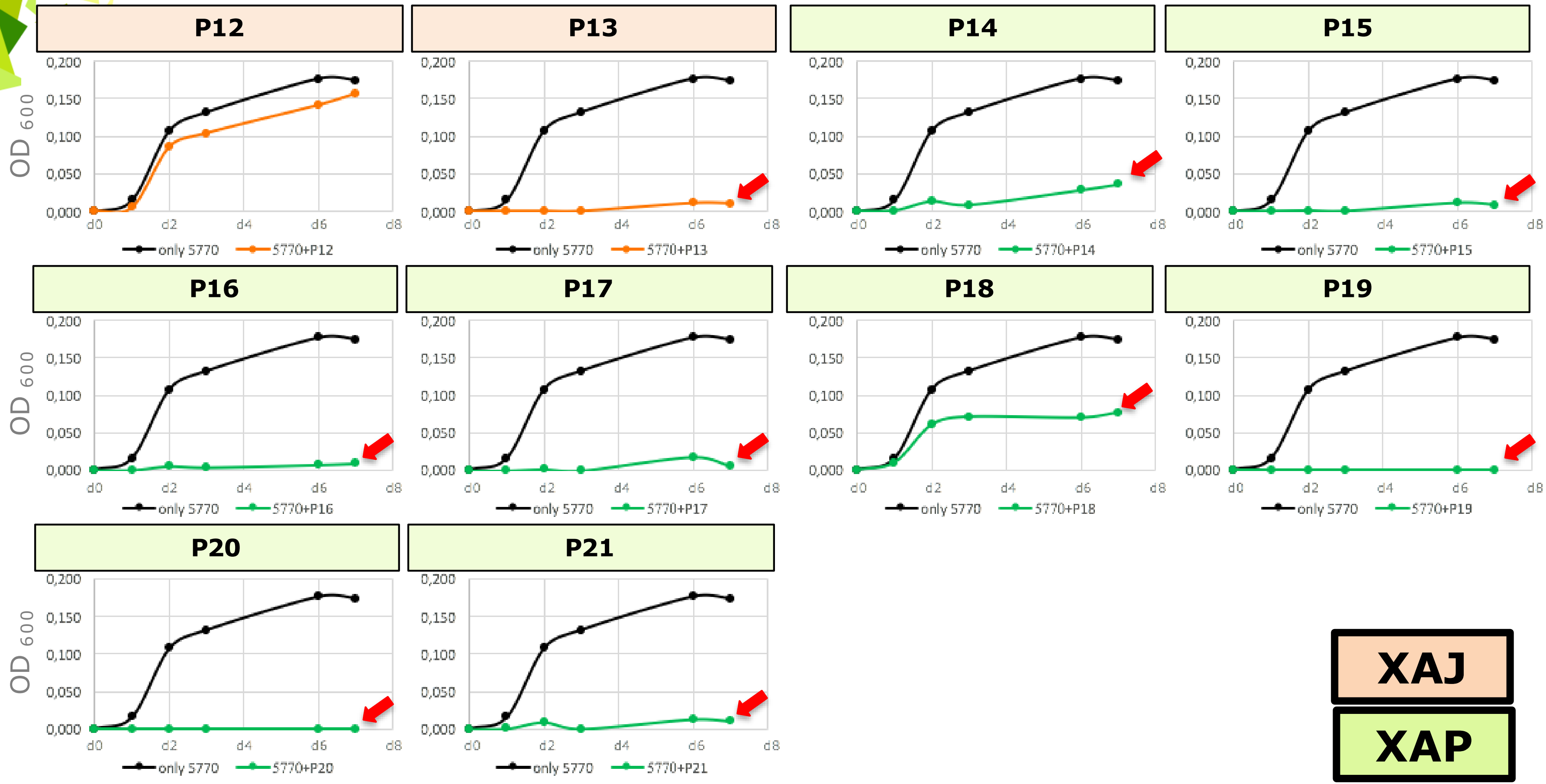
XAJ



RESULTS

X. fastidiosa subsp. *fastidiosa* **IVIA 5770**

Host range



XAJ

XAP

RESULTS

Specie	subespecie	Strain	ST	Host	Origin	Year
Xylella fastidiosa	*fastidiosa*	Temecula 1	ST 1	Grapevine	California (USA)	1998
	fastidiosa	IVIA 5235	ST 1	Cherry	Mallorca (Spain)	2016
	fastidiosa	IVIA 5388	ST 1	Almond	Mallorca (Spain)	2017
	fastidiosa	IVIA 5770	ST 1	Grapevine	Mallorca (Spain)	2017
	fastidiosa	IVIA 5772	ST 1	Grapevine	Mallorca (Spain)	2017
	fastidiosa	IVIA 6015	ST 1	*Rhamnus alaternus*	Mallorca (Spain)	2017

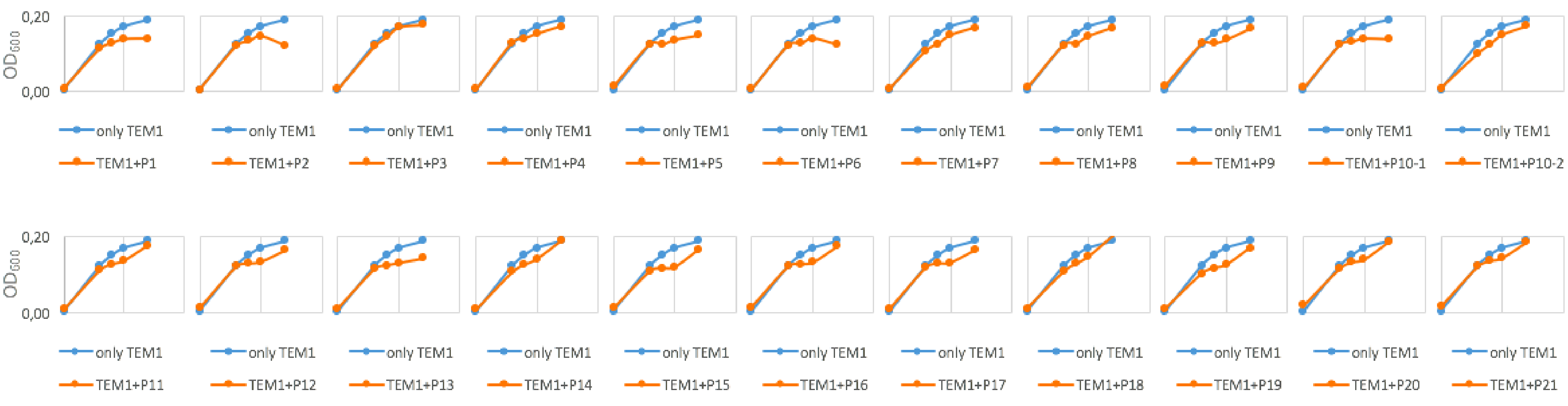
RESULTS

Host range

Specie	subespecie	Strain	ST	Host	Origin	Year
Xylella fastidiosa	*fastidiosa*	Temecula 1	ST 1	Grapevine	California (USA)	1998
	fastidiosa	IVIA 5235	ST 1	Cherry	Mallorca (Spain)	2016
	fastidiosa	IVIA 5388	ST 1	Almond	Mallorca (Spain)	2017
	fastidiosa	IVIA 5770	ST 1	Grapevine	Mallorca (Spain)	2017
	fastidiosa	IVIA 5772	ST 1	Grapevine	Mallorca (Spain)	2017
	fastidiosa	IVIA 6015	ST 1	*Rhamnus alaternus*	Mallorca (Spain)	2017

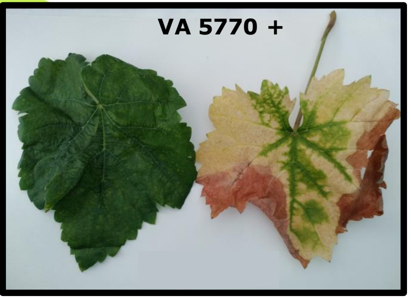
Strains susceptible to viral infection

Temecula1



RESULTS

ACTIVITY AGAINST *Xylella fastidiosa*



Strain: IVIA 5770

Subespecie: fastidiosa

ST: 1

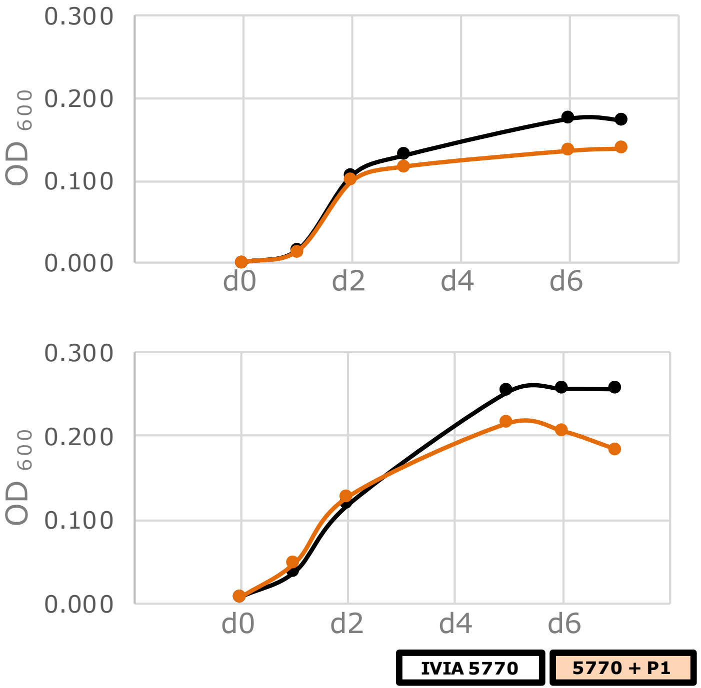
Host: grape

Origin: Mallorca-Spain

Year: 2017

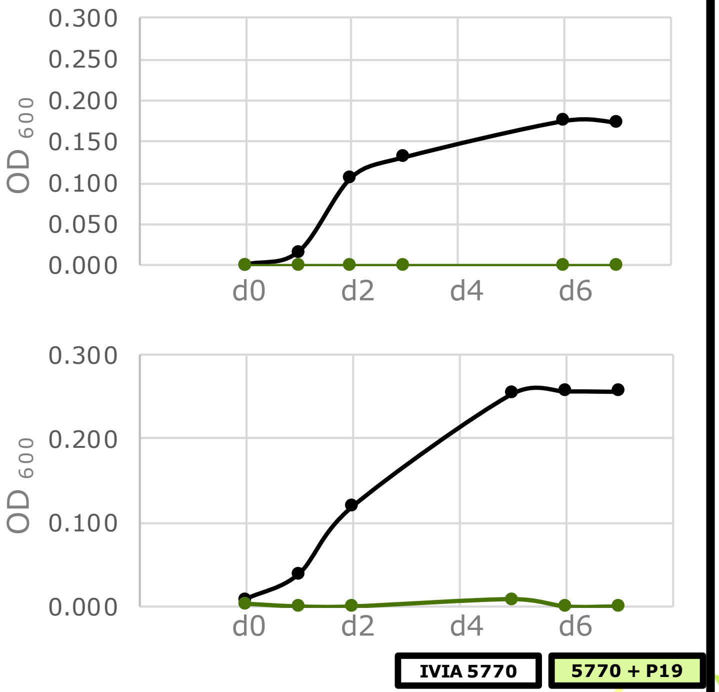
XAJ

P1



XAP

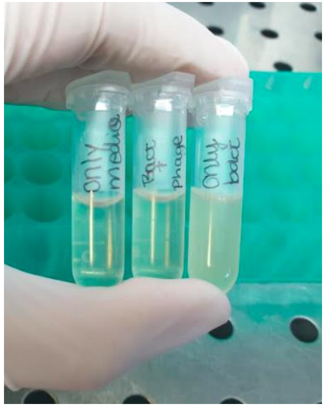
P19



RESULTS

PHAGE ACTIVITY AGAINST *Xylella fastidiosa*

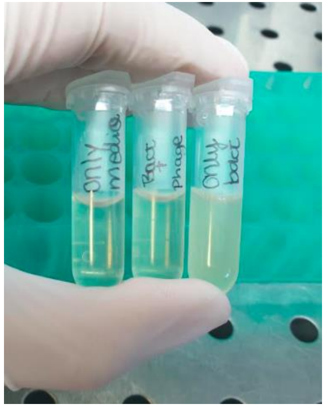
Phage P13

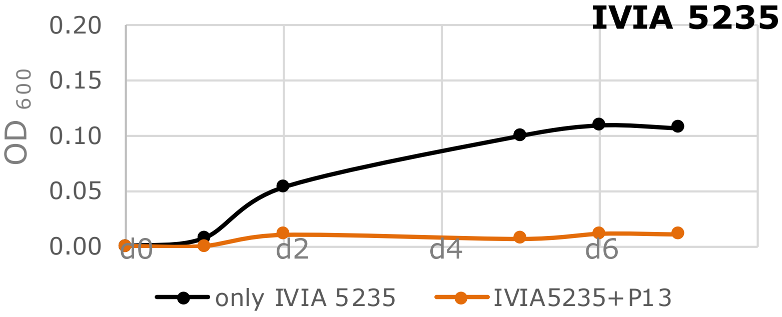


RESULTS

PHAGE ACTIVITY AGAINST *Xylella fastidiosa*

Phage P13

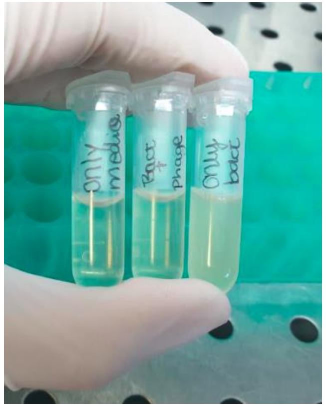


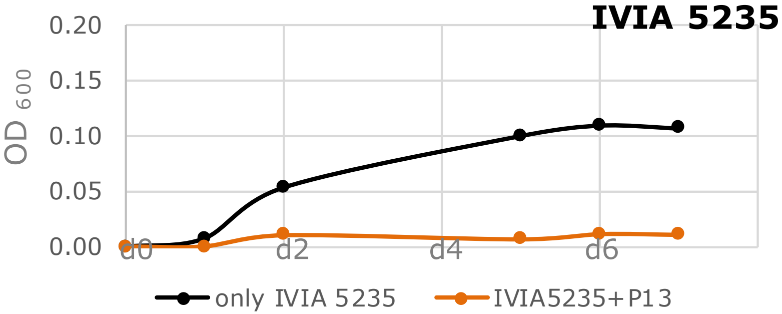


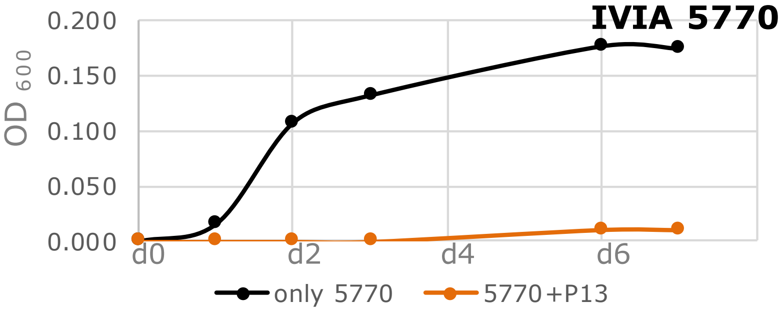
RESULTS

PHAGE ACTIVITY AGAINST *Xylella fastidiosa*

Phage P13



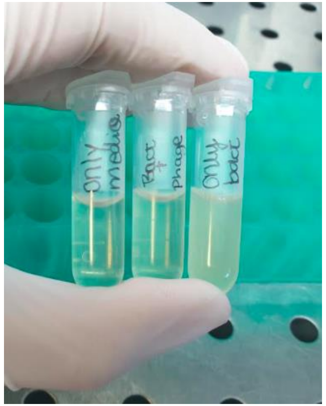


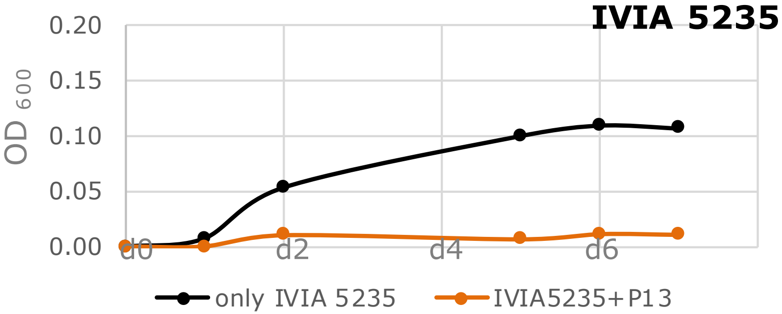


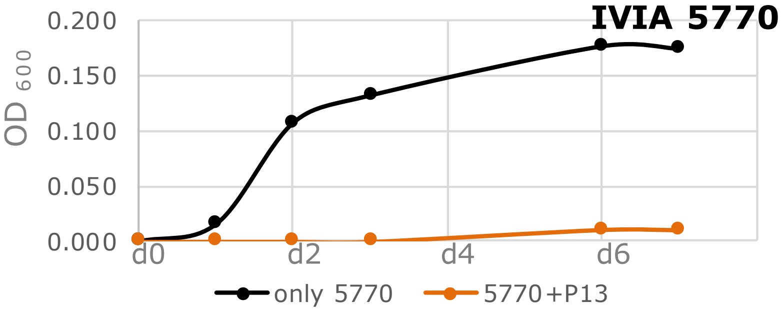
RESULTS

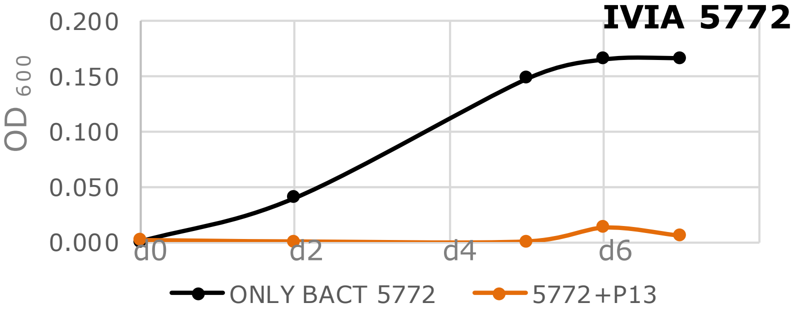
PHAGE ACTIVITY AGAINST *Xylella fastidiosa*

Phage P13





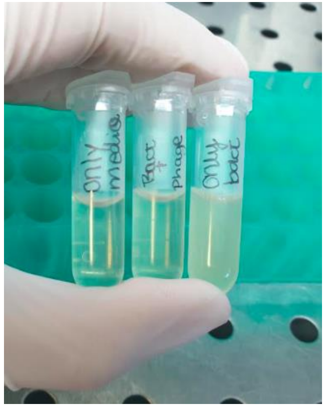


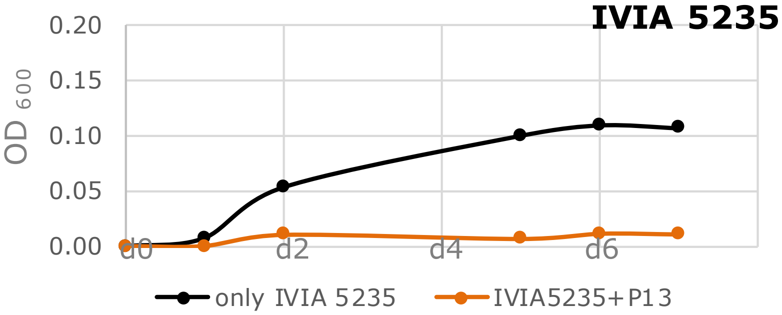


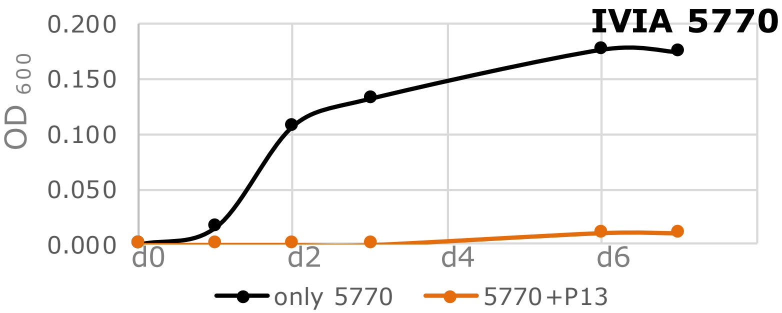
RESULTS

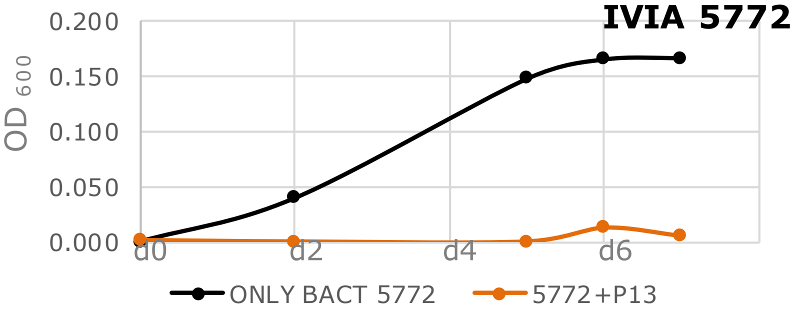
PHAGE ACTIVITY AGAINST *Xylella fastidiosa*

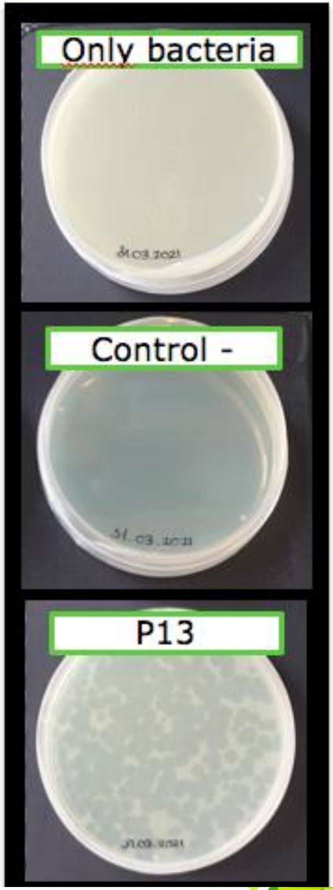
Phage P13







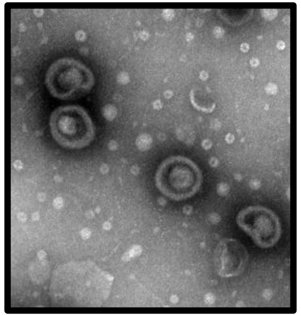


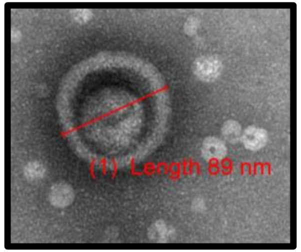


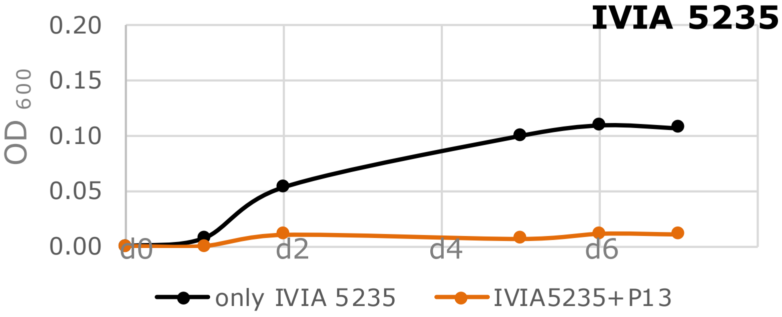
RESULTS

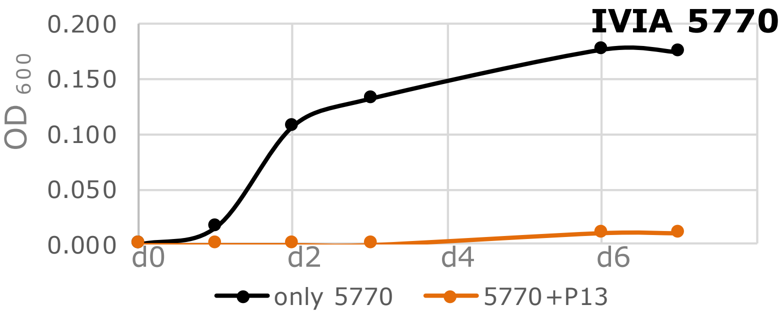
PHAGE ACTIVITY AGAINST *Xylella fastidiosa*

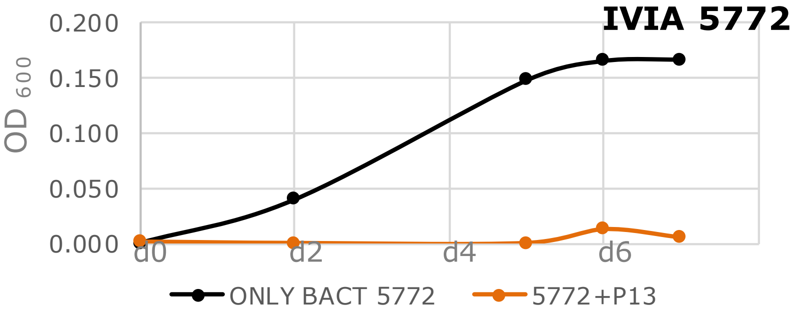
Phage P13

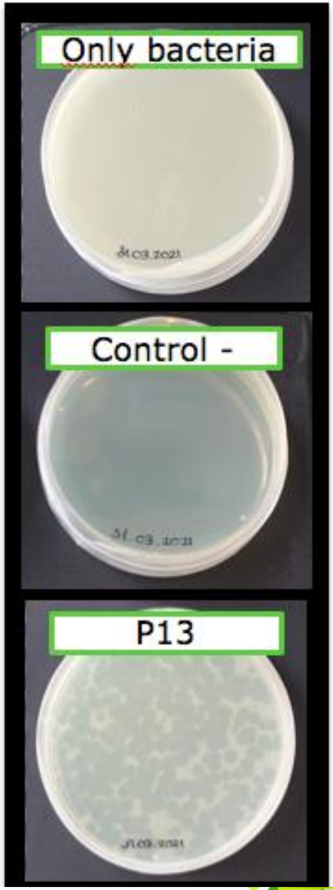












RESULTS

Phage purification, sequencing and genomic characterization

XAJ

X. arboricola pv. *juglandis* (IVIA 1317-1a)

PHAGE	
P1	P8
P2	P9
P3	P10-1
P4	P10-2
P5	P11
P6	P12
P7	P13

XAP

X. axonopodis pv. *phaseoli* (CECT 914)

PHAGE
P14
P15
P16
P17
P18
P19
P20
P21

RESULTS

Phage purification, sequencing and genomic characterization

XAJ

X. arboricola pv. *juglandis* (IVIA 1317-1a)

PHAGE	
P1	P8
P2	P9
P3	P10-1
P4	P10-2
P5	P11
P6	P12
P7	P13

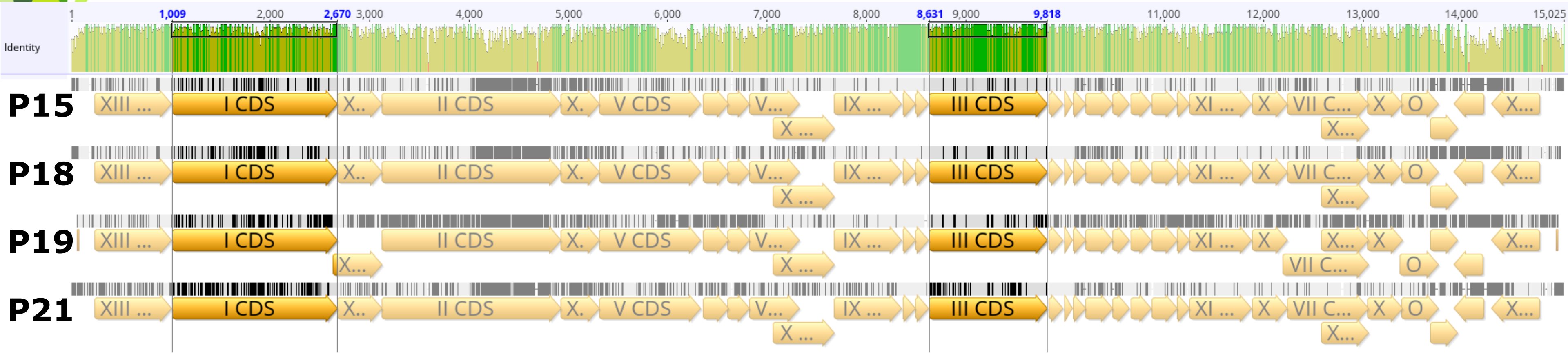
XAP

X. axonopodis pv. *phaseoli* (CECT 914)

PHAGE
P14
P15
P16
P17
P18
P19
P20
P21

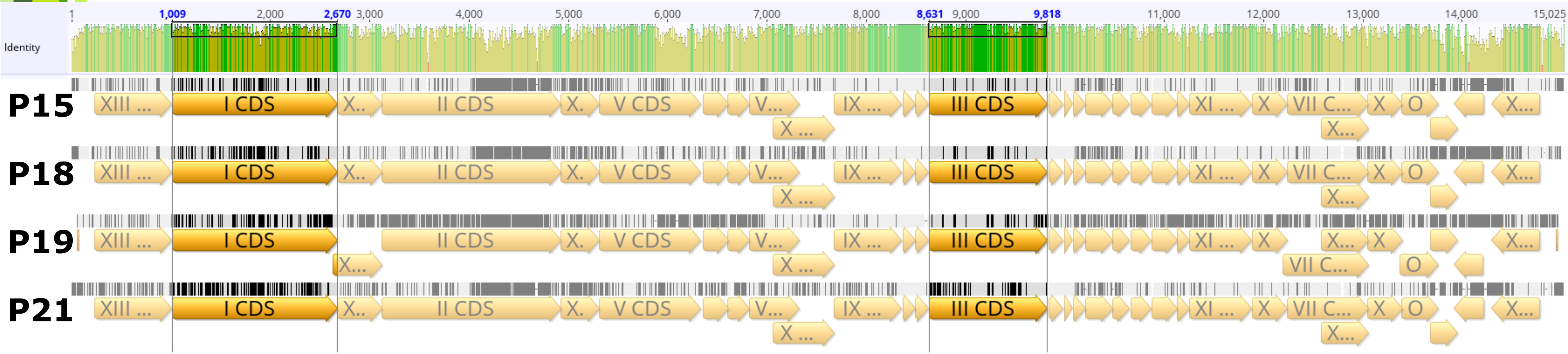
RESULTS

Phage sequencing and genomic characterization

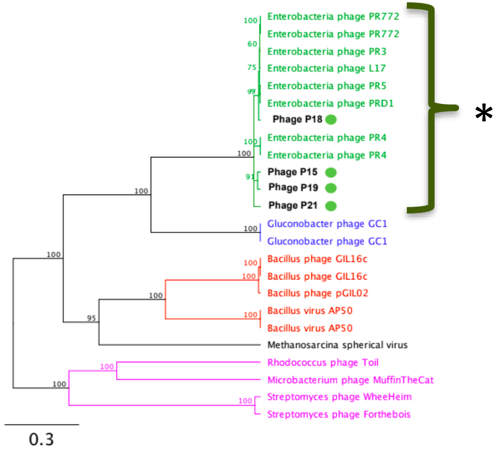


RESULTS

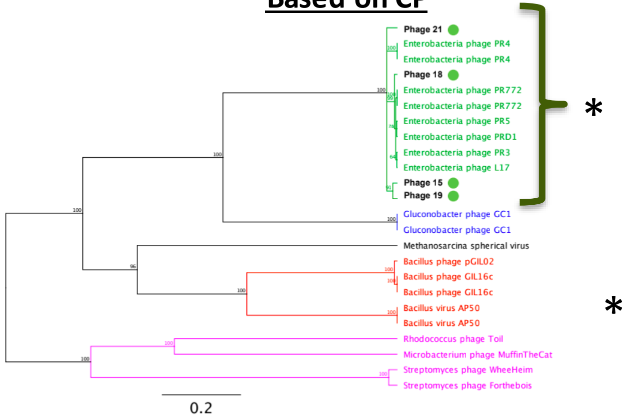
Phage sequencing and genomic characterization



Based on DNA polymerase



Based on CP



* **Family:** *Tectiviridae*
Genus: Alphatectivirus

The evolutionary distances were computed using the p-distance method and are in the units of the number of amino acid differences per site.

RESULTS

Electron transmission microscopy

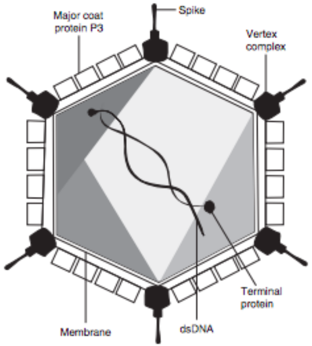


Figure 1 Schematic presentation of PRD1 virion.

Family: *Tectiviridae*
Genus: Alphatectivirus

RESULTS

Electron transmission microscopy

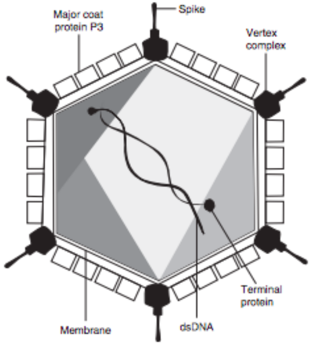
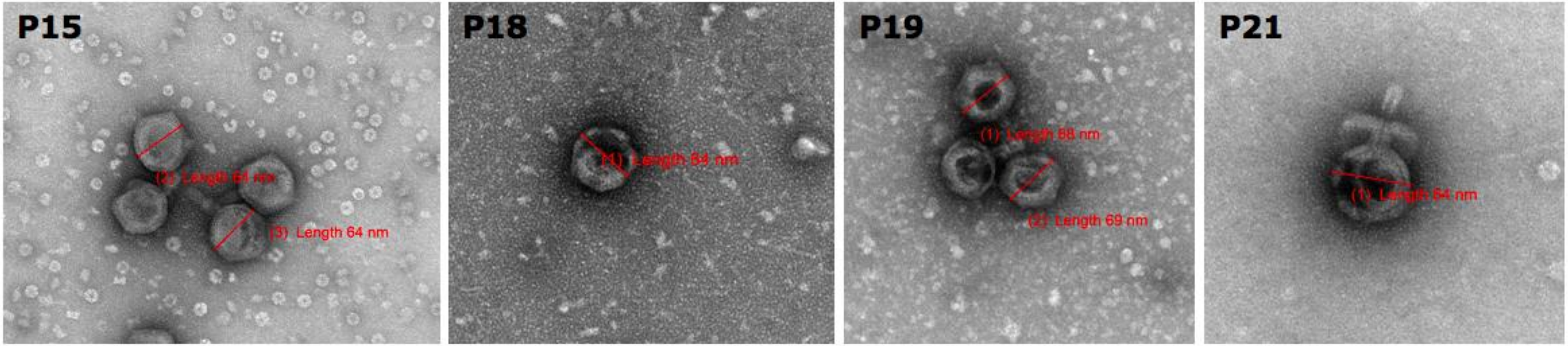
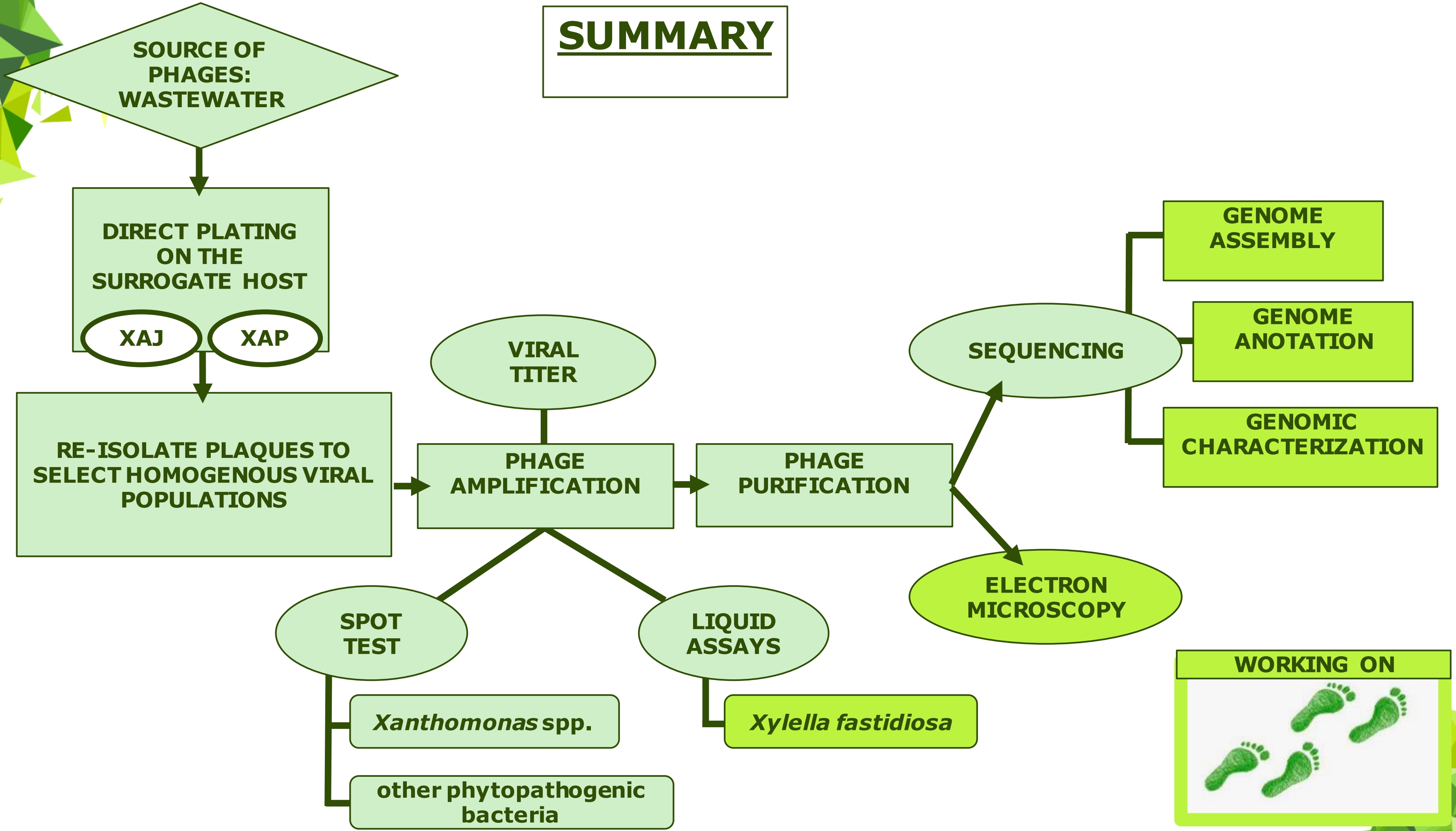


Figure 1 Schematic presentation of PRD1 virion.

Family: *Tectiviridae*
Genus: Alphatectivirus

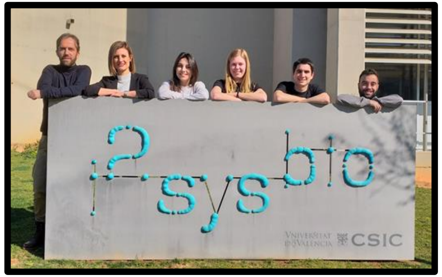


SUMMARY


SOURCE OF PHAGES: WASTEWATER
DIRECT PLATING ON THE SURROGATE HOST
XAJ
XAP
RE-ISOLATE PLAQUES TO SELECT HOMOGENOUS VIRAL POPULATIONS
VIRAL TITER
PHAGE AMPLIFICATION
SPOT TEST
Xanthomonas spp.
other phytopathogenic bacteria
LIQUID ASSAYS
Xylella fastidiosa
PHAGE PURIFICATION
SEQUENCING
GENOME ASSEMBLY
GENOME ANOTATION
GENOMIC CHARACTERIZATION
ELECTRON MICROSCOPY
WORKING ON

ACKNOWLEDGMENTS

Dr. Ester Marco-Noales
Grupo Bacterias fitopatógenas - IVIA



Dr. Pilar Domingo Calap
Enviromental and biomedical viruses (EnBiVirLab)
I2SYSBIO (UV-CSIC)

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ivia Institut Valencià d'Investigacions Agràries
GrupoTragsa
VNIVERSITAT DE VALÈNCIA
i2sysbio INSTITUTE FOR INTEGRATIVE SYSTEMS BIOLOGY
CSIC CONSEJO SUPERIOR DE INVESTIGACIONES CIENTÍFICAS

efsa
European Food Safety Authority

XF ACTORS

3rd European Conference on Xylella fastidiosa and XF-ACTORS final meeting

Isolation and characterization of bacteriophages against *Xylella fastidiosa*

M.L. Domingo-Calap, C.M. Aure, I. Navarro-Herrero, P. Domingo-Calap, E. Marco-Noales

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