



***Xylella fastidiosa* subsp. *pauca* and *multiplex* elicit differential responses in a susceptible olive cultivar**

Annalisa Giampetruzzi

Istituto per la Protezione Sostenibile delle Piante (CNR), Italy



3rd European
conference on
***Xylella*
*fastidiosa***
2021

**3rd EFSA European Conference on *Xylella fastidiosa* and
final meeting of the XF-ACTORS project**
26-30 April 2021



Olive trees (cultivated and wild) infected by different genotypes of *X. fastidiosa* subsp. *pauca* and *multiplex* in Europe

ST81 subsp. *multiplex*
(Mallorca & Menorca)
wild olive

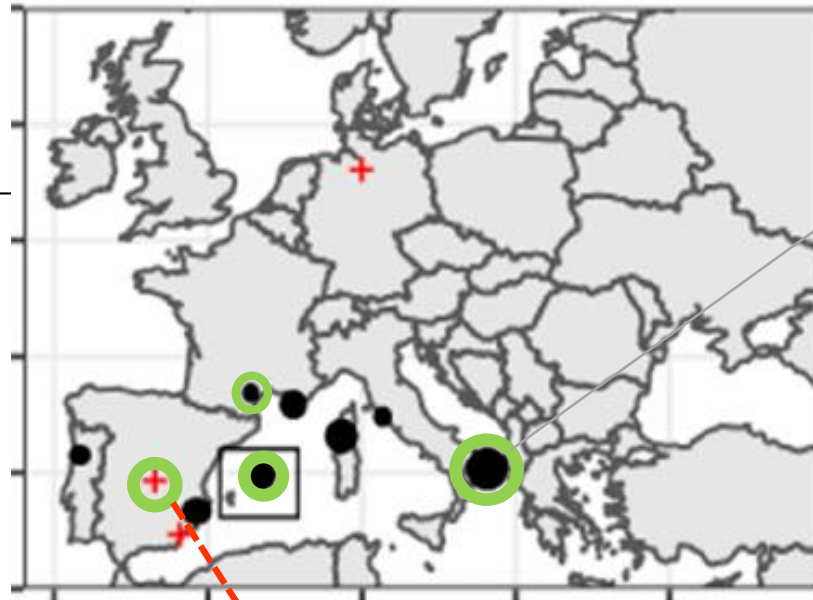


ST80 subsp. *pauca*
(Ibiza)



Olives infected by *X. fastidiosa* in the **Balearic Islands** (updated to December 2020) :

Species	Xf Subspecies	ST	Number of Samples qPCR+ ¹	Number of Samples Analyzed by qPCR
<i>Olea europaea europaea</i>	<i>multiplex</i> & <i>pauca</i>	ST80 & ST81	193	1975
<i>Olea europaea sylvestris</i>	<i>multiplex</i> & <i>pauca</i>	ST80 & ST81	495	3012



1 report in France (ST53)

Update of the Xylella spp. host plant database up to 30 June 2019. EFSA J. 2020, 18, e06114

ST53 subsp. *pauca* (Apulia, Italy)



*Prunus dulcis***

?



Olea europaea

- + = Outbreaks considered eradicated
- = areas where Xf is well established
- = Xf outbreak in olive

ST6 outbreak on olive in the
Community of Madrid (Spain)
(April 2018) >>> **ERADICATED**

Xf subsp. *multiplex* **ST6** was mainly associated to
almond leaf scorch disease (ALSD) in **Alicante**
(Valencian Community-Spain), where it **has not been**
detected causing infection in olive trees.

**Could olive potentially become a
host of ST6 genotype?**

Do olive plants inoculated by ST6 strains show symptoms ?

(Nunney et al., 2019)

Pathogenicity experiments in olive (Spain)

X. fastidiosa pauca (ST53, ST80) and *X. fastidiosa multiplex* (ST6, ST81)

Olive Genotypes

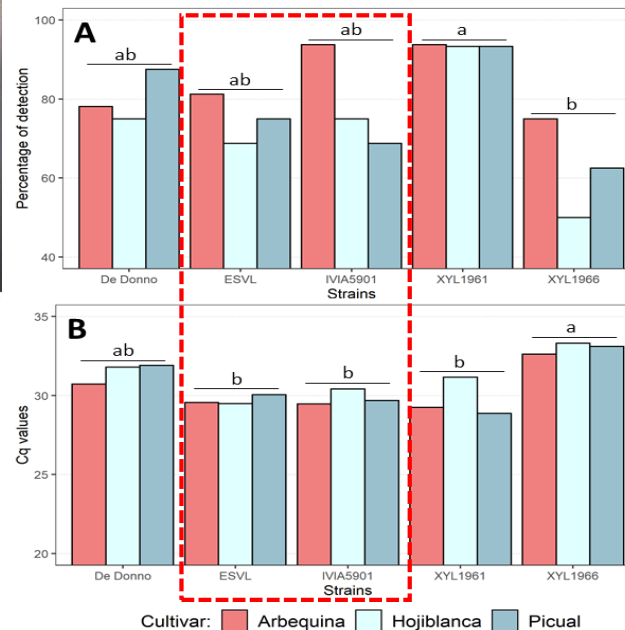
Picual, Hojiblanca y Arbequina



Experiment	Strain	Subspecie	Isolated from	Geographic origin	ST
A	IVIA5901	<i>multiplex</i>	<i>Prunus dulcis</i>	Bol lulla, Alicante (Spain)	6
A	ESVL	<i>multiplex</i>	<i>Prunus dulcis</i>	Benimantel, Alicante (Spain)	6
B	XYL1966	<i>multiplex</i>	<i>Olea europea</i>	Mallorca, Islas Baleares (Spain)	81
B	XYL1961	<i>pauca</i>	<i>Olea europea</i>	Ibiza, Islas Baleares (Spain)	80
A and B	De Donno	<i>pauca</i>	<i>Olea europea</i>	Lecce (Italy)	53



Low concentration of bacteria



**no symptoms
after more than 2 years on
the inoculated olives**

(poster from Miguel Roman et al., 2021
3rd EFSA European Conference)

Blanca B. Landa

An Experimental Test of the Host-Plant Range of Nonrecombinant Strains of North American *Xylella fastidiosa* subsp. *multiplex*

Leonard Nunney,^{1,*} Hamid Azad,² and Richard Stouthamer³

TABLE 2. Leaf symptoms (mean $\pm$ standard error) and the percentage of the stem infected as measured by enzyme-linked immunosorbent assay (ELISA) in all plant hosts successfully infected by at least one *Xylella fastidiosa* subsp. *multiplex* strain^a

Plant host	Symptoms and infected (%) for <i>X. fastidiosa</i> subsp. <i>multiplex</i> strain, name, sequence type (ST)							
	ALS003	ALS299	OLV029	PLP070	PLP069	OAK309	LIQ062	LIQ064
	Dixon	M12	G12	Pc053	Pc052	2689OAK	LS020	LS043
	ST6	ST7	ST7	ST26	ST36	ST9	ST24	ST24
Almond	4.9 $\pm$ 0.1 (85) 4.8 $\pm$ 0.1(100)	2.7 $\pm$ 0.6 (90) 4.7 $\pm$ 0.2 (95)	0.0 $\pm$ 0.0 (0)	3.3 $\pm$ 0.3 (100) 4.2 $\pm$ 0.4 (100)	0.0 $\pm$ 0.0 (0)	0.0 $\pm$ 0.0 (0)	0.0 $\pm$ 0.0 (0)	3.6 $\pm$ 0.1 (100) 2.3 $\pm$ 0.2 (76)
Plum	3.0 $\pm$ 0.5 (74) 2.5 $\pm$ 0.3 (48)	3.2 $\pm$ 0.6 (76)	0.0 $\pm$ 0.0 (0)	4.3 $\pm$ 0.2 (100) 4.0 $\pm$ 0.3 (100) 4.2 $\pm$ 0.2 (84)	4.2 $\pm$ 0.4 (100)	0.0 $\pm$ 0.0 (0)	4.0 $\pm$ 0.3 (93) 2.5 $\pm$ 0.3 (68)	0.0 $\pm$ 0.0 (0)
Peach	0.0 $\pm$ 0.0 (0)	0.0 $\pm$ 0.0 (0)	0.0 $\pm$ 0.0 (0)	3.3 $\pm$ 0.3 (100) 3.7 $\pm$ 0.4 (92)	0.0 $\pm$ 0.0 (0)	0.0 $\pm$ 0.0 (0)	0.0 $\pm$ 0.0 (6)	0.0 $\pm$ na (0)
Blackberry	...	2.3 $\pm$ 0.2 (52)	...	2.8 $\pm$ 0.2 (56)	...	2.8 $\pm$ 0.2 (65)	...	0.0 $\pm$ 0.0 (19) ^d
Red oak	0.0 $\pm$ 0.0 (0)	0.0 $\pm$ na (13)	0.0 $\pm$ 0.0 (6)	0.0 $\pm$ na (0) 1.0 $\pm$ na (67) ^b	0.0 $\pm$ 0.0 (0)	1.0 $\pm$ na (75) ^c	0.0 $\pm$ 0.0 (0) 1.0 $\pm$ na (50) ^b	0.0 $\pm$ na (0)
Grapevine	2.2 $\pm$ 0.3 (30) 1.9 $\pm$ 0.3 (37) 2.5 $\pm$ 0.3 (41)	0.0 $\pm$ 0.0 (0) 0.0 $\pm$ 0.0 (0)	2.5 $\pm$ 0.3 (84)	0.0 $\pm$ 0.0 (0) 0.0 $\pm$ 0.0 (0)	0.0 $\pm$ 0.0 (0)	0.0 $\pm$ 0.0 (0)	0.0 $\pm$ 0.0 (12) 0.0 $\pm$ 0.0 (0)	0.0 $\pm$ 0.0 (0)
Sweetgum	0.0 $\pm$ 0.0 (0)	0.0 $\pm$ 0.0 (0)	...	0.0 $\pm$ 0.0 (0)	...	0.0 $\pm$ 0.0 (0)	3.0 $\pm$ 0.3 (82)	3.5 $\pm$ 0.3 (79)
Olive	0.0 $\pm$ 0.0 (20)	0.0 $\pm$ 0.0 (0)	2.5 $\pm$ 0.3 (86)	0.0 $\pm$ 0.0 (0)	0.0 $\pm$ 0.0 (0)	0.0 $\pm$ 0.0 (0)	0.0 $\pm$ 0.0 (17)	...

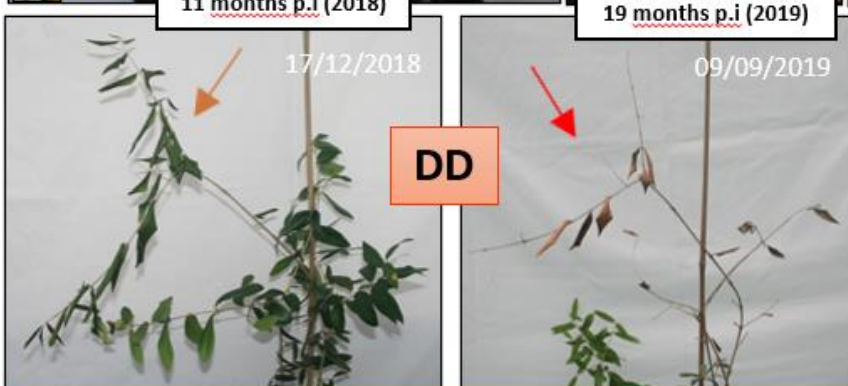
No leaf symptoms on olive inoculated by ALS003 (ST6):

Which is the olive response to Xf (ST6) in very susceptible cultivar as Cellina?

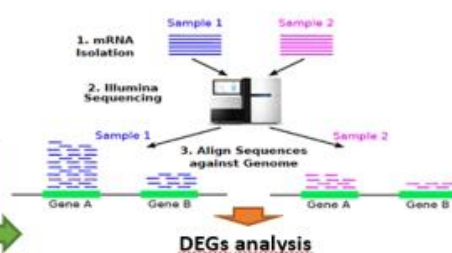


Pathogenicity experiments in olive (Italy) with ESVL strain (ST6)

cv.Cellina di Nardò (greenhouse condition)



Olives infected by De Donno (ST53) showed severe desiccation symptoms 18 months p.i. as expected, some plants are dead at 3 years p.i.



Transcriptomic study on these olive plants (18 months p.i.)



Evolution of symptoms



Maria Saponari,
Giuseppe Altamura

Quantification of bacteria

(2 years p.i.):

- De Donno: Cq of 23 (average)
- ESLV Cq of 28 (average)

(3 year p.i.)

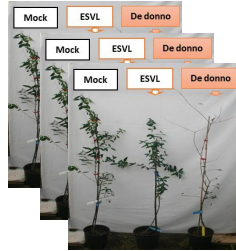


Olives infected by ESVL (ST6) showed non-specific symptoms 3 year p.i. !

Exploring the host response to *Xf* multiplex (ST6) by transcriptomic approach

Experimental design:

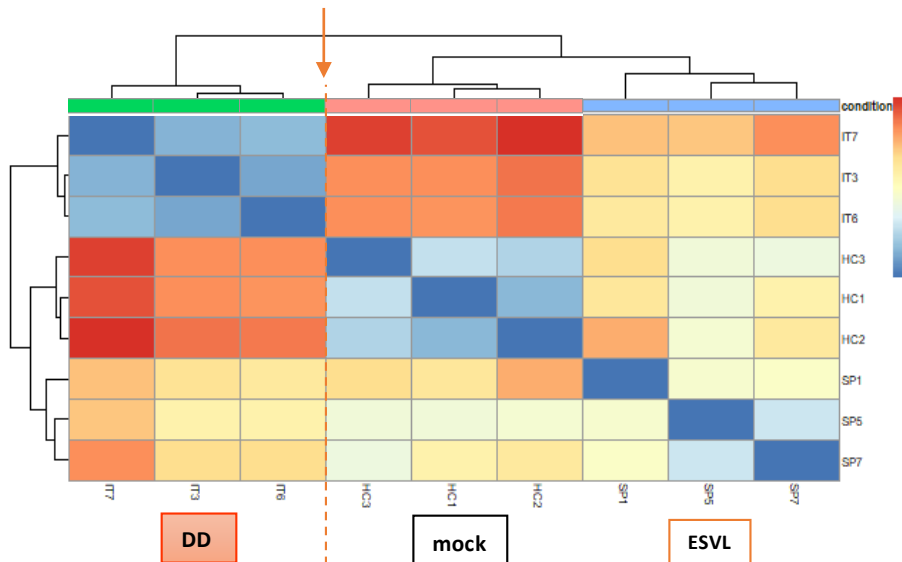
Biol.Replicates:	Rep1	Rep2	Rep3	Rep1	Rep2	Rep3	Rep1	Rep2	Rep3
Sample ID	HC1	HC2	HC3	SP1	SP5	SP7	IT3	IT6	IT7
cultivar	CELLINA	CELLINA	CELLINA	CELLINA	CELLINA	CELLINA	CELLINA	CELLINA	CELLINA
condition/ <i>Xf</i> strain	Mock/Hty	Mock/Hty	Mock/Hty	Xf_ESVL	Xf_ESVL	Xf_ESVL	Xf_DeDonno	Xf_DeDonno	Xf_DeDonno



Statistic of mapping on the three reference olive genomes:

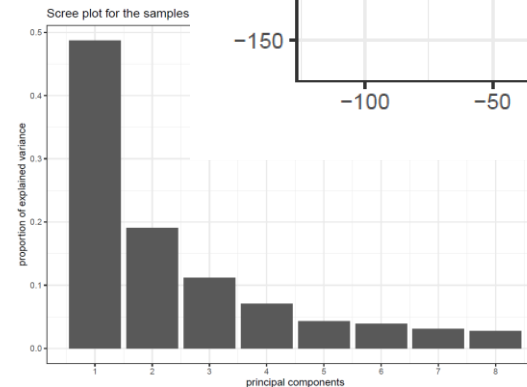
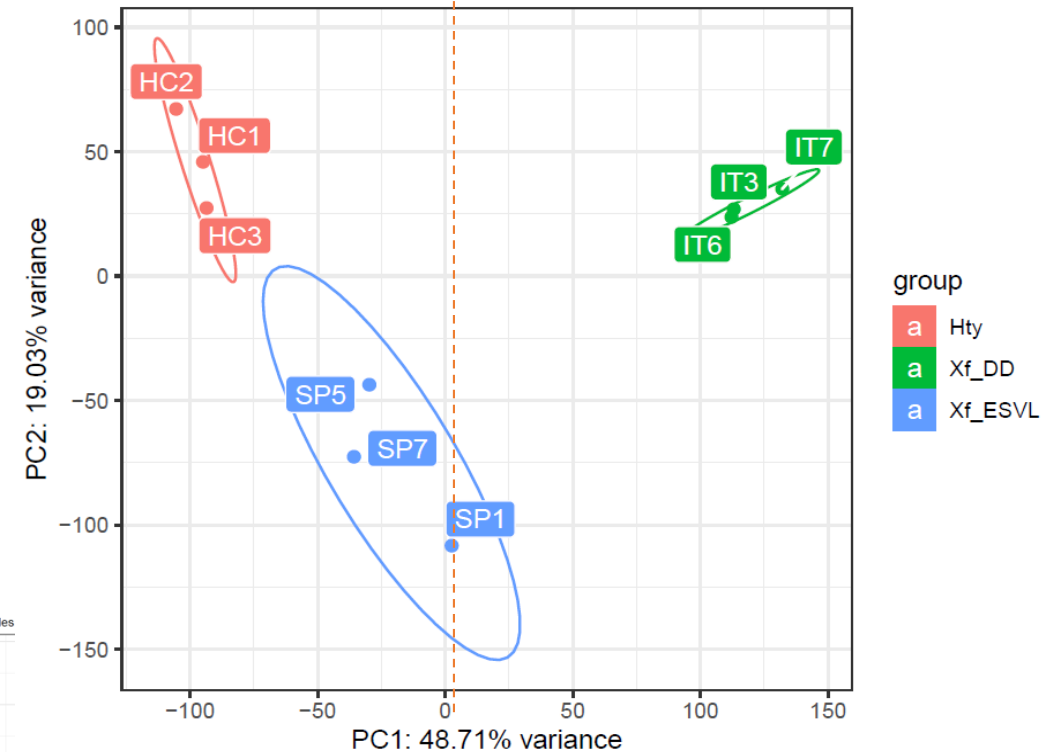
reference olive genomes:			cv. <i>Farga</i> (Oea6)			var. <i>sylvestris</i>		cv. <i>Picual</i>	
SAMPLE ID	cultivar	condition	raw reads	mapped reads	rate of mapping (%)	mapped reads	rate of mapping (%)	mapped reads	rate of mapping (%)
HC1	CELLINA	Hty	39,229,432	31,159,951	79.4	27,441,705	70	32,202,602	82.1
HC2	CELLINA	Hty	43,975,112	35,226,853	80.1	30,500,992	69.4	36,486,930	83
HC3	CELLINA	Hty	44,199,422	36,380,331	82.3	31,788,092	71.9	37,515,110	84.9
IT3	CELLINA	DD	37,102,126	30,026,477	80.9	26,532,899	71.5	31,029,554	83.6
IT6	CELLINA	DD	44,213,778	36,197,153	81.9	31,872,659	72.1	37,400,876	84.6
IT7	CELLINA	DD	41,087,208	33,160,102	80.7	29,191,924	71	34,282,691	83.4
SP1	CELLINA	ESVL	33,698,590	27,626,856	82	24,395,663	72.4	28,533,834	84.7
SP5	CELLINA	ESVL	43,740,218	35,039,553	80.1	31,024,754	70.9	36,263,261	82.9
SP7	CELLINA	ESVL	41,531,896	33,563,043	80.8	29,462,808	70.9	34,803,220	83.8

Countdatasets of 79,867 annotated transcripts x 9 samples



Sample to sample distance analysis first separate mock and ESVL from DD samples

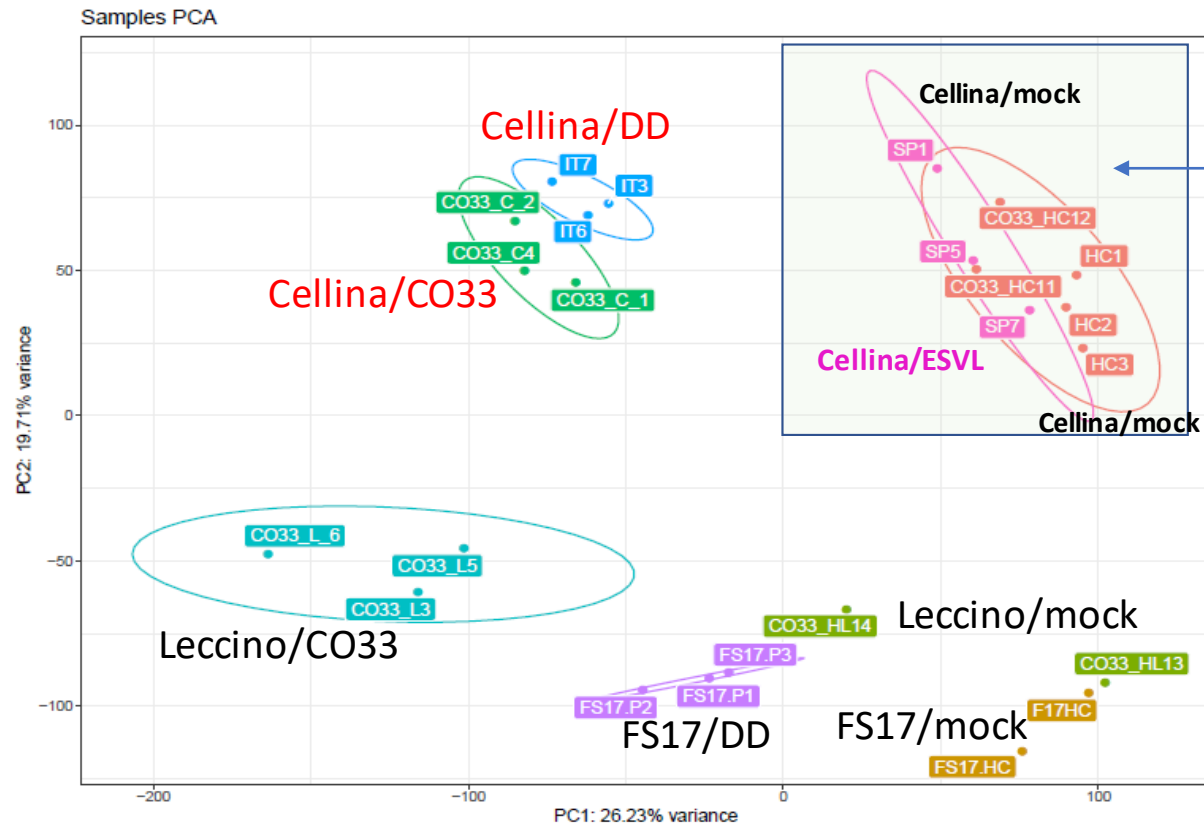
Principal Component Analysis (PCA) on samples



PCA clearly grouped olive infected by **ESVL** close to **mock/healthy** samples according the PC1 which explain the 48% of total variability

ESVL samples together with all RNAseq samples (artificial inoculation trials)

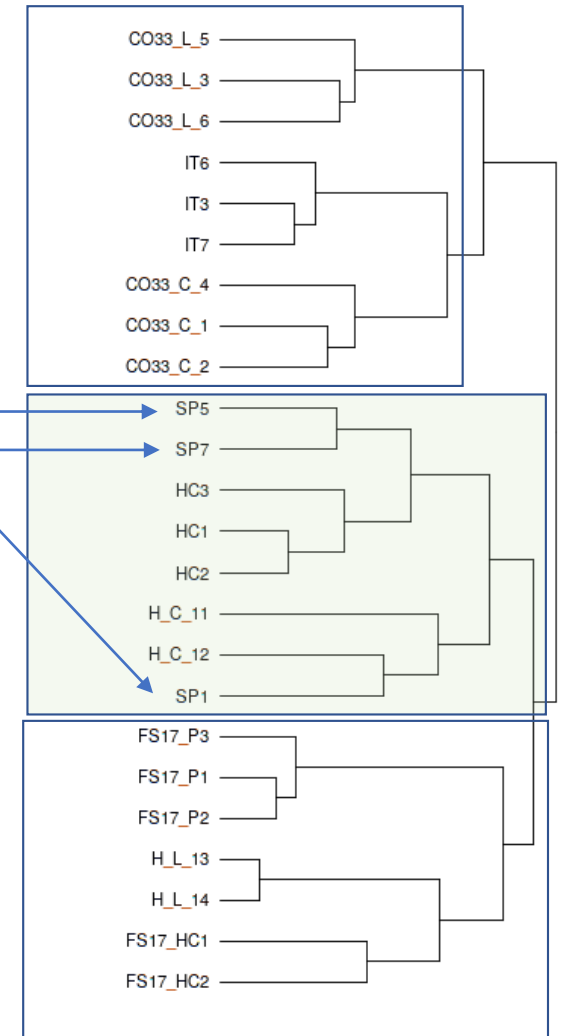
Trial	Biological replicates:	Rep1	Rep2	Rep3	Rep1	Rep2	Rep1	Rep2	Rep3	Rep1	Rep2
1	Sample ID	CO33_C_1	CO33_C_2	CO33_C_4	H_C_11	H_C_12	CO33_L_3	CO33_L_5	CO33_L_6	H_L_13	H_L_14
	cultivar	CELLINA	CELLINA	CELLINA	CELLINA	CELLINA	LECCINO	LECCINO	LECCINO	LECCINO	LECCINO
	condition/Xf strain	Xf_CO33	Xf_CO33	Xf_CO33	Mock/Hty	Mock/Hty	Xf_CO33	Xf_CO33	Xf_CO33	Mock/Hty	Mock/Hty
2	Sample ID	IT3	IT6	IT7	HC1	HC2	HC3	SP1	SP5	SP7	
	cultivar	CELLINA	CELLINA	CELLINA	CELLINA	CELLINA	CELLINA	CELLINA	CELLINA	CELLINA	
	condition/Xf strain	Xf_DeDonno	Xf_DeDonno	Xf_DeDonno	Mock/Hty	Mock/Hty	Mock/Hty	Xf_ESVL	Xf_ESVL	Xf_ESVL	
2	sample_name	FS17-P1	FS17-P2	FS17-P3	F17HC	FS17-HC					
	cultivar	FS17	FS17	FS17	FS17	FS17					
	condition/Xf strain	Xf_DeDonno	Xf_DeDonno	Xf_DeDonno	Mock/Hty	Mock/Hty					



Olive infected by ESVL (ST6) were grouped together with healthy/mock cv. Cellina samples

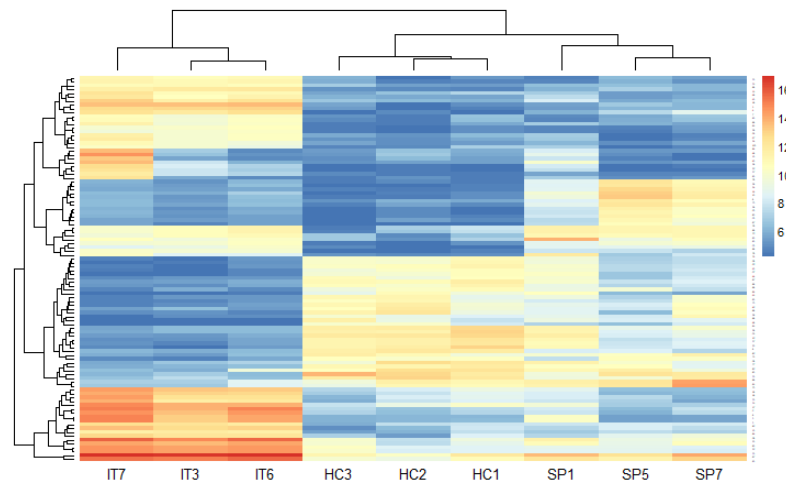
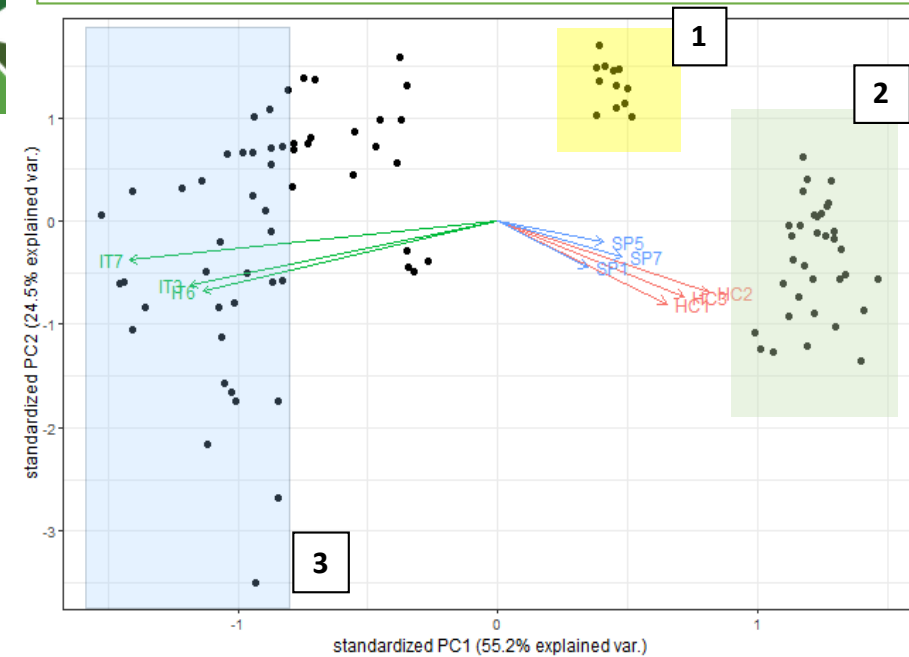
Correlation (average linkage)

0.0 0.2 0.4 0.6 0.8 1.0 1.2

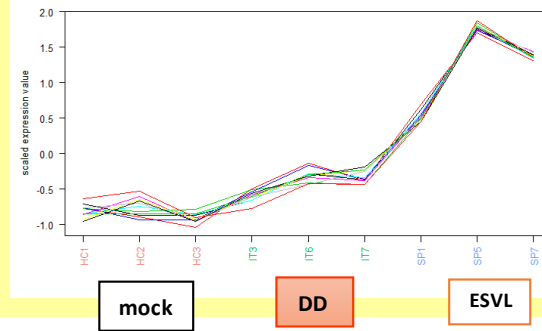


Exploring the host response to Xf multiplex (ST6) by transcriptomic approach:

PCA and Hierarchical clustering on most 100 variable transcripts



Group
 → Hty
 → Xf_DD
 → Xf_ESVL

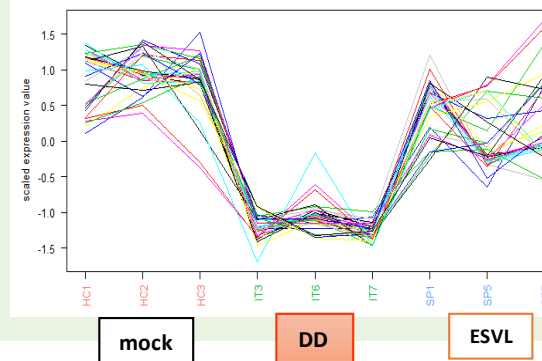


"Expansin-like B1"

are carbohydrate-binding protein involved in cell-wall loosening (Zhang et al., 2014)
 Upregulation of Exp B is positively associated with drought tolerance

"Bidirectional sugar transporter SWEET3"

Xanthomonas species are able to upregulate the transcription of SWEET transporters in rice (Oryza sativa) by secretion of TAL effectors.



Cellulose synthase A catalytic subunit 4

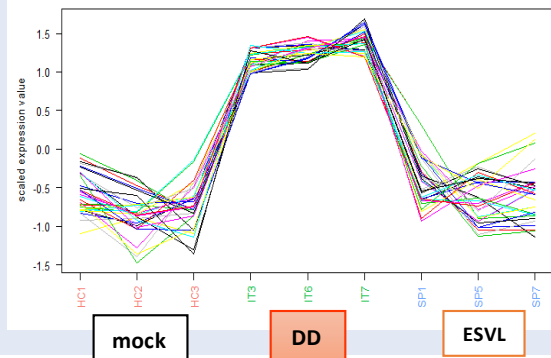
Low expression when secondary wall synthesis takes place.

Fascidin-like arabinogalactan protein 11

Expressed in the sclerenchyma cells of inflorescence stems

Leucine-rich repeat extensin-like protein 3

Expressed mostly in vascular tissues, modulates cell morphogenesis by regulating cell wall formation and assembly



LRR receptor-like serine/threonine-protein kinase

PR proteins (Thaumatin, PR6, PR1, PR3 endochitinase)

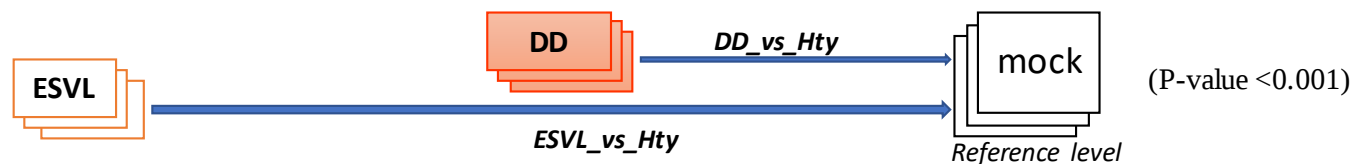
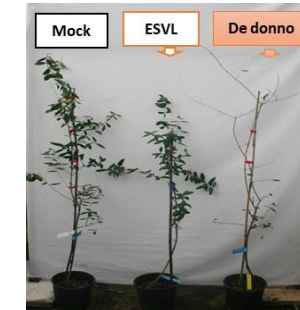
Peroxidase

Cannabidiolic acid synthase

oxidases that inactivate the elicitor-active oligogalacturonides (Benedetti et al 2018)

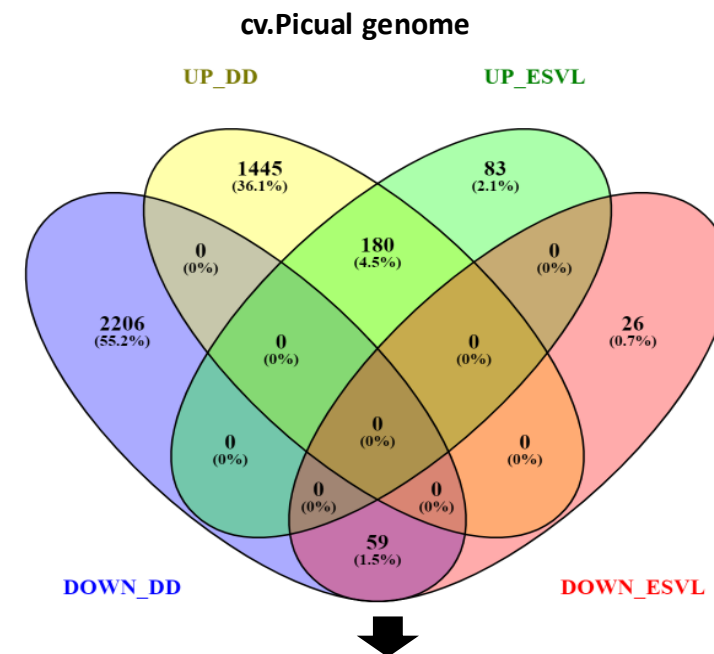
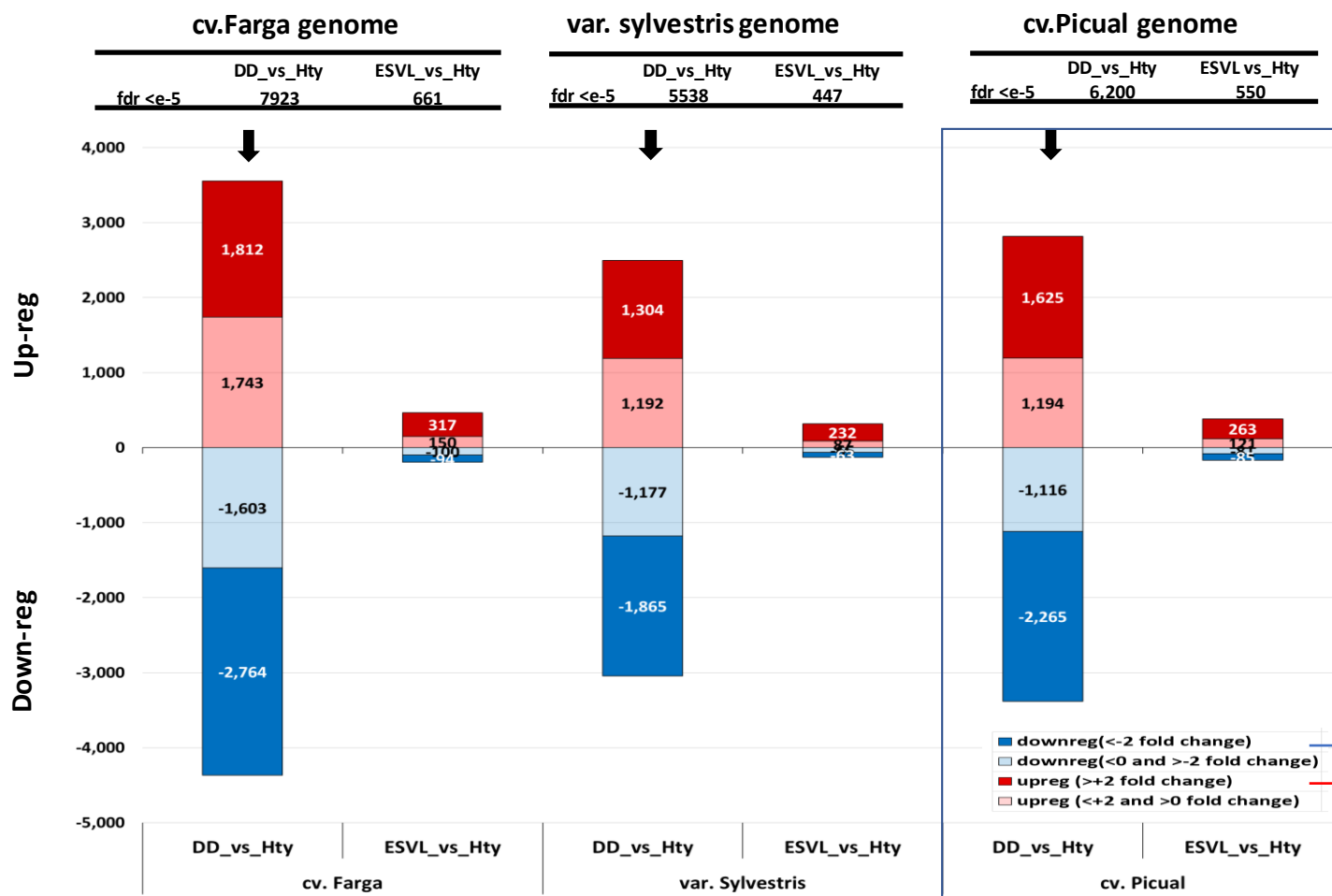
**DEGs NOT
upregulated in
olive infected
by ESVL SP
samples**

Differential Gene Expression (DEG) analysis (DESeq2, Venny tools)



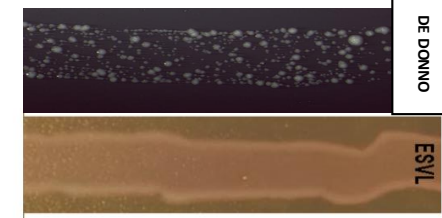
Much higher number of DEGs in response to infection by De Donno compared to infection by ESVL !

in **cv. Farga countdataset** we found an higher number of DEGs (FDR<0.00001), but more than **33%** (i.e. 2416/7923) of **DEGs transcripts lack functional annotation**

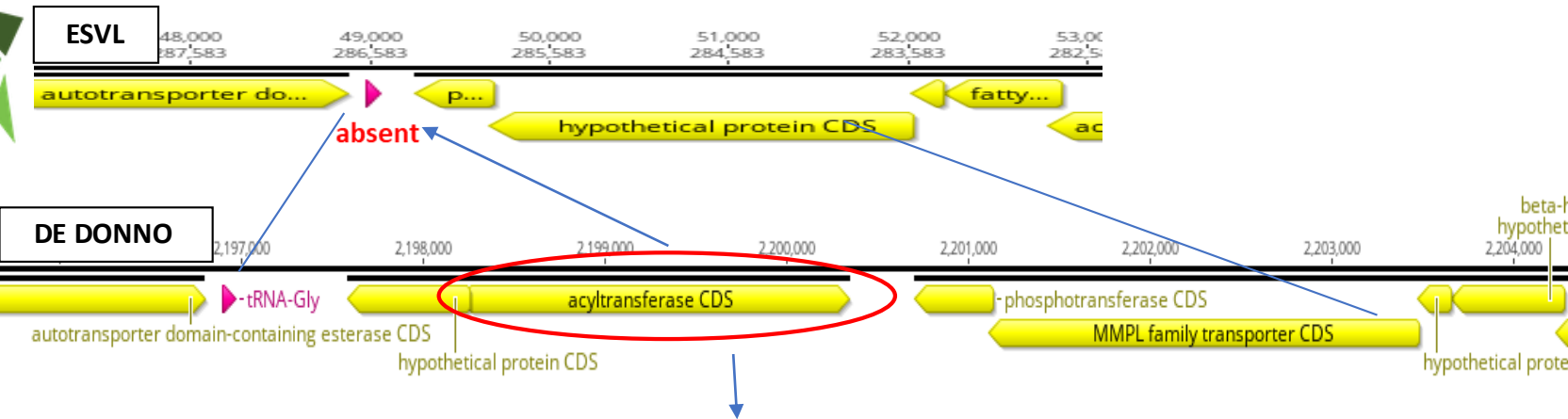


Gene Ontology (GO) enrichment using the singular enrichment analysis (SEA) with agriGO v2

Are there differences in bacterial genomes?: De donno vs ESVL



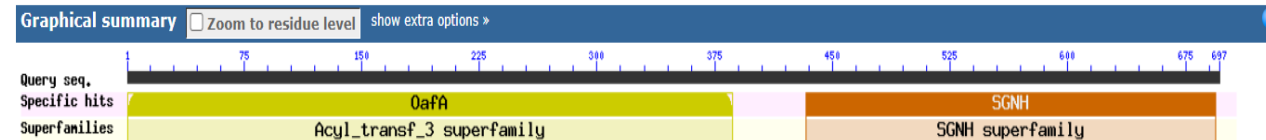
Acyltransferase or “O-antigen acetylase gene”



South American supsp. *pauca* strain (9a5c) contained a gene encoding an O-antigen acetylase (**OafA**), which is absent in all North American strains (Van Sluys et al 2003)

OafA is involved in the acylation of carbohydrate moieties on extracellular molecules (LPS) (Barbosa et al., 2015)

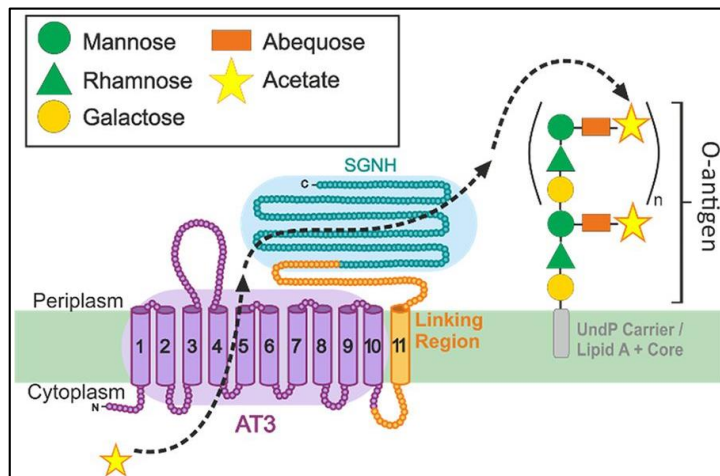
acyltransferase [Xylella fastidiosa] **CCD domain searching >>> two domains (AT3, SGNH)**



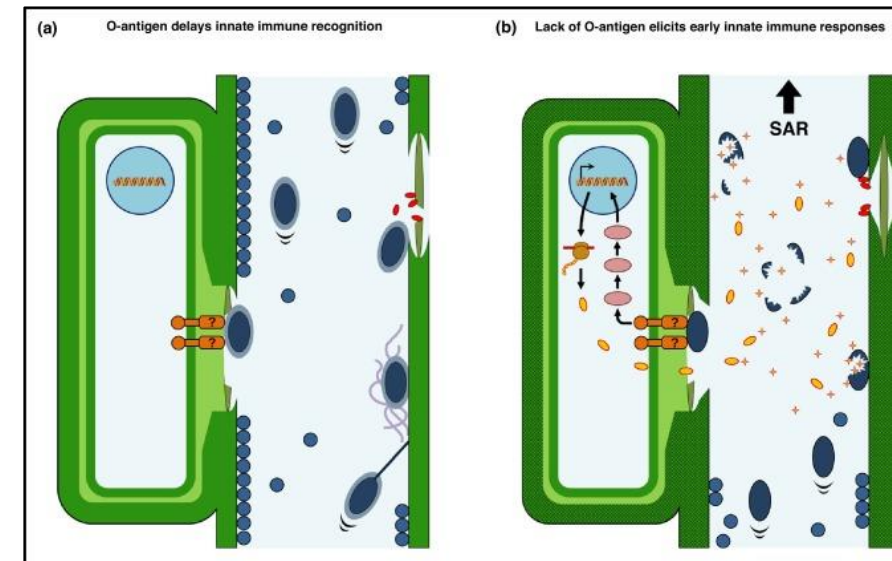
Lack of the O-antigen modifies plant perception of Xf and enables elicitation of hallmarks of PTI, specifically in the plant xylem tissue in grape (Rapicavoli et al 2018)

Proposed mechanism of action of O antigen acetyltransferases during maturation of the LPS in the periplasm of **Salmonella enterica** using OafA

affecting immunogenicity, virulence, and bacteriophage resistance (Pearson et al., 2020)

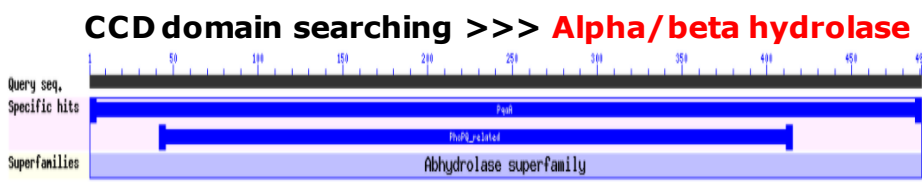
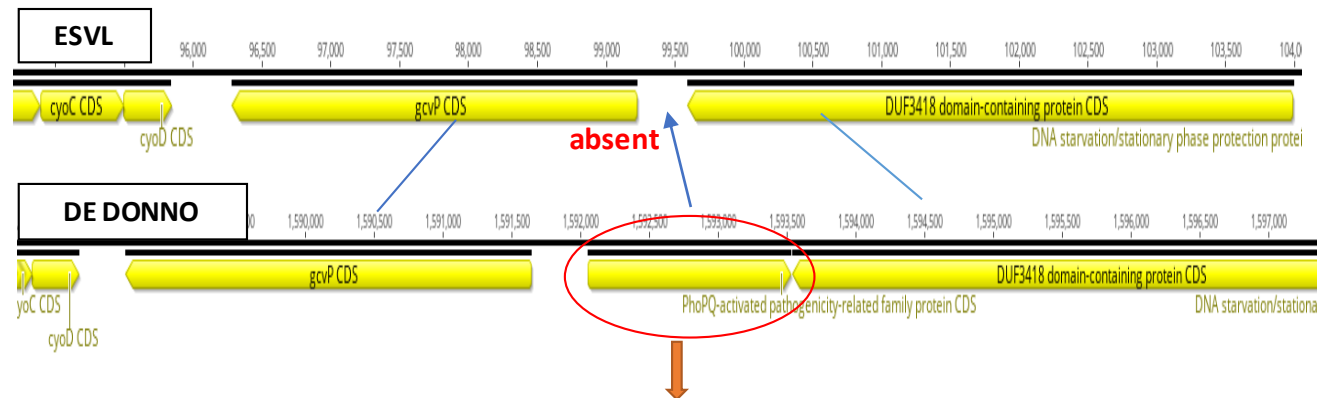


Could **lacking of gene** for **acylation of LPS** structures act as determinants for host specificity and/or virulence amongst Xf bacteria?



Differences in genomic composition: ESVL vs De donno

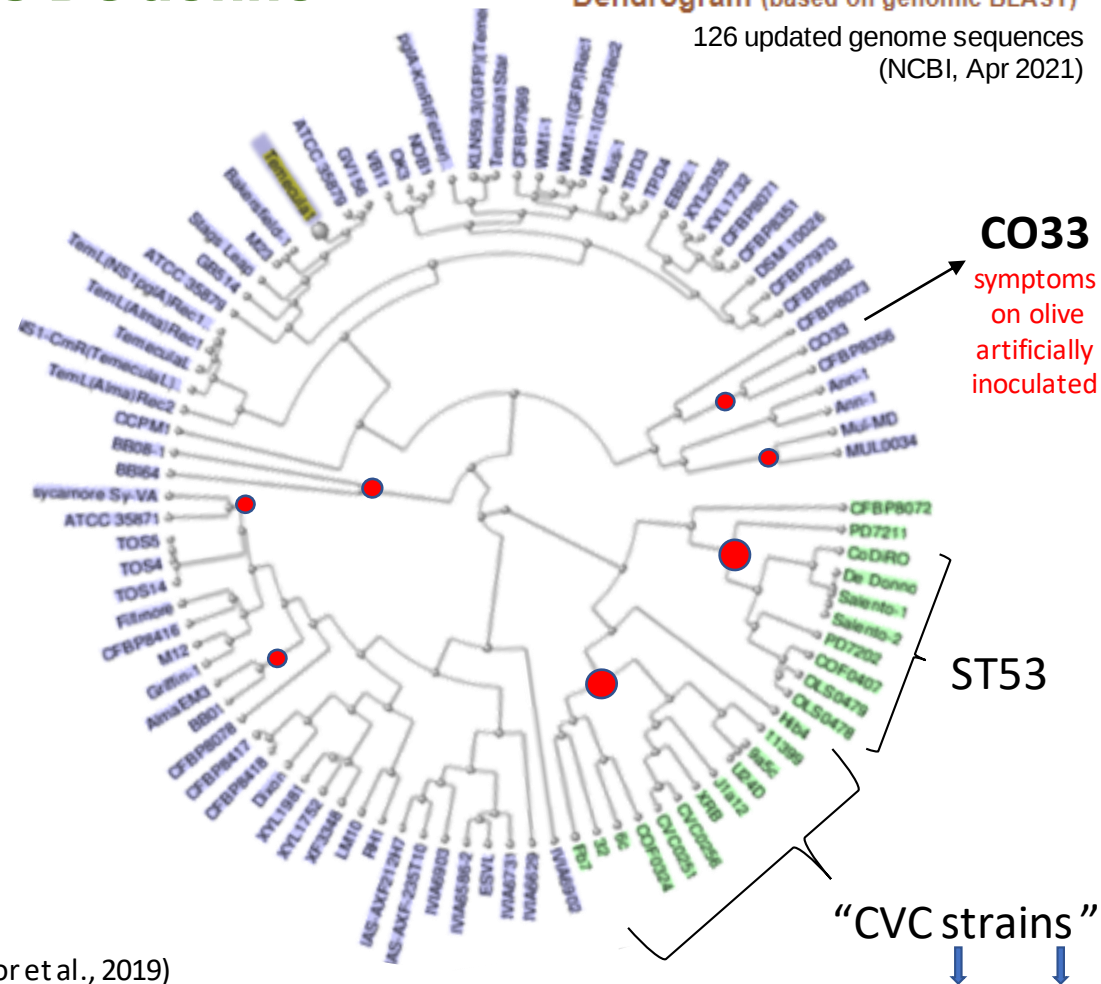
gene coding for
“PhoPQ-activated pathogenicity-related family protein, PqA” ●



A functionally diverse superfamily containing proteases, lipases, peroxidases, esterases, epoxide hydrolases and dehalogenases

Dendrogram (based on genomic BLAST)

126 updated genome sequences
(NCBI, Apr 2021)



PqA in OR Feitosa-Junior et al., 2019)

TABLE 1. Top abundant proteins identified in the outer membrane vesicle (OMV)-enriched preparations from *Xylella fastidiosa* strains Temecula1, 9a5c, and Fb7

CDS number _{CVC}	CDS number _{PD} ^a	CDS symbol	Gene product	Ranking (scaffold score)		
				Temecula1	9a5c	Fb7
XF0357	PD1702	LesA	Lipase/Esterase LesA	1	14	7
XF0358	PD1703	LesB	Lipase/Esterase LesB	2	24	11
XF1384	–	PqA	Alpha/beta-Hydrolases PqA	–	57	18
XF2551	PD1934		Dipeptidyl aminopeptidase protein 6	–	64	34
XF1024	PD0312		Membrane lipoprotein	–	–	25
XF1745	–		Alpha/beta-Hydrolases	–	–	31

Question to answer:

“Which is the involvement of **PqA** protein”
in the *Xf* pathogenicity in olive ?

CONCLUSIONS

- **Olive** is one of the **host species** infected in **different European Xf-outbreaks** mainly by subsp. *pauca* (**ST53 in Italy**) and ST80 in Ibiza, but new ST (subsp. *multiplex*) are detected on olive: ST81 in Mallorca&Menorca, **ST6** in Mainland Spain (eradicated)
- **ESVL strain (ST6)** was mainly associated to **almond causing ALSD**, but pathogenicity test were started **to test if olive is a host for this strain**
- Olive **plants** artificially inoculated with **ESVL strain (ST6)** have **not developed any symptoms** after three years, whereas olive plants inoculated with **De Donno strain (ST53)** showed 100% incidence of symptoms and **severe desiccation** (including death of some plant)
- **Transcriptome from olives (cv. Cellina)** artificially infected by **De donno strain (ST53)** as expected was much more **drastically reprogrammed to activate defense response** against the bacteria compared to the transcriptome from olives infected by ESLV strain (ST6)
- The **comparative genomic and transcriptomic approach** used in our study was useful to **identify defense responses** in olive plants and to identify potentially genes that interact with *X. fastidiosa* providing **useful insights for exploring the molecular basis of Xylella-olive interaction**

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Blanca B. Landa,



Ester Marco Noales

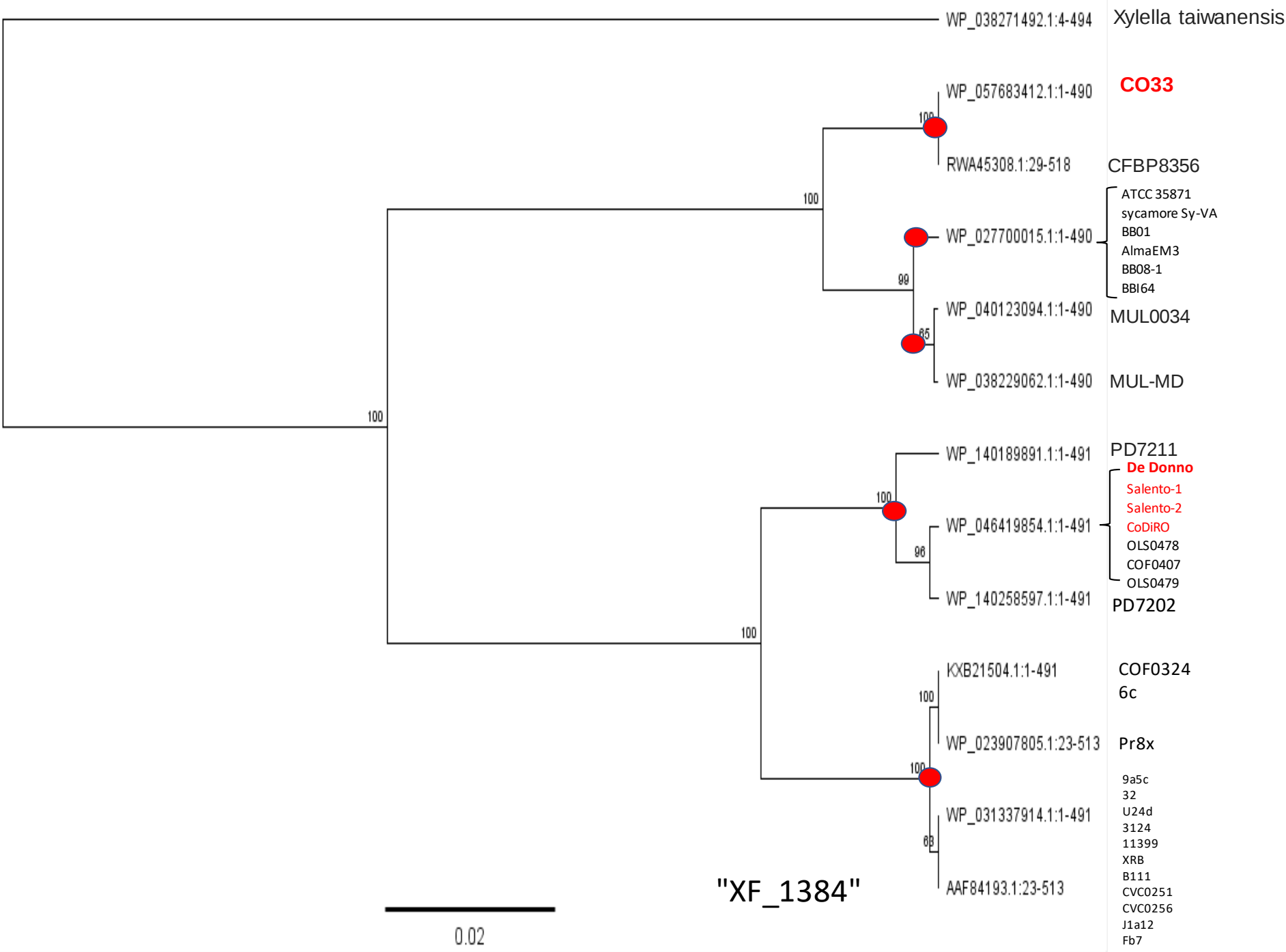


"A journey from resilience to resistance"

O-antigen acetylase gene

CP020870.12198251-2200344 (+)	ARO69397.1	acyltransferase	Xylella fastidiosa subsp. pauca	De Donno	GCA_002117875.1
CP016608.12198073-2200166 (+)	AVI21420.1	hypothetical protein	Xylella fastidiosa	Salento-1	GCA_002954185.1
CP016610.12198261-2200354 (+)	AVI23455.1	hypothetical protein	Xylella fastidiosa	Salento-2	GCA_002954205.1
JUJW01000002.1140528-142621 (-)	KIA58669.1	hypothetical protein	Xylella fastidiosa	CoDiRO	GCA_000811965.1
LRV101000035.1294958-297051 (-)	KXB10778.1	hypothetical protein	Xylella fastidiosa	OLS0478	GCA_001549755.1
LRVH01000109.1140361-142454 (-)	KXB16342.1	hypothetical protein	Xylella fastidiosa	OLS0479	GCA_001549735.1
LRVJ01000030.139098-41191 (+)	KXB16922.1	hypothetical protein	Xylella fastidiosa	COF0407	GCA_001549825.1
RRUA01000001.11639385-1641478 (+)	TNW26288.1	acyltransferase	Xylella fastidiosa subsp. pauca	PD7202	GCA_006370235.1
NZ_RRTZ01000001.1956700-958793 (+)	WP_140190018.1	acyltransferase	Xylella fastidiosa subsp. pauca	PD7211	GCF_006370175.1
NZ_CP009885.12461526-2463619 (+)	WP_088578403.1	acyltransferase	Xylella fastidiosa	Hib4	GCF_001456315.1
CP009826.22340457-2342700 (+)	ALR05009.2	acyltransferase	Xylella fastidiosa	Pr8x	GCA_001456295.2
NZ_AXBS02000016.139132-41309 (+)	WP_158515132.1	acyltransferase family protein	Xylella fastidiosa 6c	6c	GCF_000506905.2
NZ_LRVG01000023.1169034-171127 (-)	WP_023908087.1	acyltransferase	Xylella fastidiosa	COF0324	GCF_001549815.1
NZ_LKDK01000010.150238-52331 (+)	WP_058569493.1	acyltransferase	Xylella fastidiosa subsp. pauca	CFBP8072	GCF_001469345.1
NC_002488.3728706-730949 (-)	WP_023906984.1	acyltransferase	Xylella fastidiosa 9a5c	9a5c	GCF_000006725.1
AWYH01000039.139166-41409 (+)	ETE29801.1	acetylase	Xylella fastidiosa 32	32	GCA_000506405.1
CP009790.1728696-730789 (-)	ALQ95818.1	hypothetical protein	Xylella fastidiosa	U24D	GCA_001456275.1
CP010051.22310199-2312292 (+)	ALR09558.2	hypothetical protein	Xylella fastidiosa	Fb7	GCA_001456335.3
NZ_JNBTO1000008.147930-50023 (-)	WP_060871712.1	acyltransferase	Xylella fastidiosa subsp. pauca 11399	11399	GCF_001684415.1
NZ_JTGF01000007.1117927-120020 (+)	WP_060871712.1	acyltransferase	Xylella fastidiosa	XR8	GCF_013283695.1
NZ_JWJX01000006.139197-41290 (-)	WP_060871712.1	acyltransferase	Xylella fastidiosa	B111	GCF_013283685.2
NZ_LRVF01000002.175804-77897 (+)	WP_060871712.1	acyltransferase	Xylella fastidiosa	CVC0251	GCF_001549765.1
NZ_LRVF01000023.138985-41078 (-)	WP_060871712.1	acyltransferase	Xylella fastidiosa	CVC0256	GCF_001549745.1
CP009823.12433506-2435599 (+)	ALR03174.1	hypothetical protein	Xylella fastidiosa	J1a12	GCA_001456235.1
LRVF01000023.138985-41078 (-)	KXB16091.1	hypothetical protein	Xylella fastidiosa	CVC0256	GCA_001549745.1
LRVF01000002.175804-77897 (+)	KXB22877.1	hypothetical protein	Xylella fastidiosa	CVC0251	GCA_001549765.1
JTGF01000007.1117927-120020 (+)	NRP54872.1	hypothetical protein	Xylella fastidiosa	XR8	GCA_013283695.1
JWJX01000006.139197-41290 (-)	NRP67785.1	hypothetical protein	Xylella fastidiosa	B111	GCA_013283685.1
JNBTO1000008.147930-50023 (-)	QCA58582.1	hypothetical protein	Xylella fastidiosa subsp. pauca 11399	11399	GCA_001684415.1
NZ_CP009829.22392835-2394760 (+)	WP_157285733.1	acyltransferase family protein	Xylella fastidiosa	3124	GCF_001456195.2

PhoPQ-activated
pathogenicity-related
family protein



Olive was not successfully infected by ST6 strains

TABLE 2. Leaf symptoms (mean $\pm$ standard error) and the percentage of the stem infected as measured by enzyme-linked immunosorbent assay (ELISA) in all plant hosts successfully infected by at least one *Xylella fastidiosa* subsp. *multiplex* strain^a

Plant host	Symptoms and infected (%) for <i>X. fastidiosa</i> subsp. <i>multiplex</i> strain, name, sequence type (ST)							
	ALS003	ALS299	OLV029	PLP070	PLP069	OAK309	LIQ062	LIQ064
	Dixon	M12	G12	Pc053	Pc052	2689OAK	LS020	LS043
	ST6	ST7	ST7	ST26	ST36	ST9	ST24	ST24
Almond	<i>4.9 $\pm$ 0.1 (85)</i> <i>4.8 $\pm$ 0.1 (100)</i>	<i>2.7 $\pm$ 0.6 (90)</i> <i>4.7 $\pm$ 0.2 (95)</i>	0.0 $\pm$ 0.0 (0)	<i>3.3 $\pm$ 0.3 (100)</i> <i>4.2 $\pm$ 0.4 (100)</i>	0.0 $\pm$ 0.0 (0)	0.0 $\pm$ 0.0 (0)	0.0 $\pm$ 0.0 (0)	<i>3.6 $\pm$ 0.1 (100)</i> <i>2.3 $\pm$ 0.2 (76)</i>
Plum	<i>3.0 $\pm$ 0.5 (74)</i> <i>2.5 $\pm$ 0.3 (48)</i>	<i>3.2 $\pm$ 0.6 (76)</i>	0.0 $\pm$ 0.0 (0)	<i>4.3 $\pm$ 0.2 (100)</i> <i>4.0 $\pm$ 0.3 (100)</i> <i>4.2 $\pm$ 0.2 (84)</i>	<i>4.2 $\pm$ 0.4 (100)</i>	0.0 $\pm$ 0.0 (0)	<i>4.0 $\pm$ 0.3 (93)</i> <i>2.5 $\pm$ 0.3 (68)</i>	0.0 $\pm$ 0.0 (0)
Peach	0.0 $\pm$ 0.0 (0)	0.0 $\pm$ 0.0 (0)	0.0 $\pm$ 0.0 (0)	<i>3.3 $\pm$ 0.3 (100)</i> <i>3.7 $\pm$ 0.4 (92)</i>	0.0 $\pm$ 0.0 (0)	0.0 $\pm$ 0.0 (0)	0.0 $\pm$ 0.0 (6)	0.0 $\pm$ na (0)
Blackberry	...	<i>2.3 $\pm$ 0.2 (52)</i>	...	<i>2.8 $\pm$ 0.2 (56)</i>	...	<i>2.8 $\pm$ 0.2 (65)</i>	...	0.0 $\pm$ 0.0 (19) ^d
Red oak	0.0 $\pm$ 0.0 (0)	0.0 $\pm$ na (13)	0.0 $\pm$ 0.0 (6)	0.0 $\pm$ na (0) 1.0 $\pm$ na (67) ^b	0.0 $\pm$ 0.0 (0)	<i>1.0 $\pm$ na (75)</i> ^c	0.0 $\pm$ 0.0 (0) 1.0 $\pm$ na (50) ^b	0.0 $\pm$ na (0)
Grapevine	<i>2.2 $\pm$ 0.3 (30)</i> <i>1.9 $\pm$ 0.3 (37)</i> <i>2.5 $\pm$ 0.3 (41)</i>	0.0 $\pm$ 0.0 (0) 0.0 $\pm$ 0.0 (0)	<i>2.5 $\pm$ 0.3 (84)</i>	0.0 $\pm$ 0.0 (0) 0.0 $\pm$ 0.0 (0)	0.0 $\pm$ 0.0 (0)	0.0 $\pm$ 0.0 (0)	0.0 $\pm$ 0.0 (12) 0.0 $\pm$ 0.0 (0)	0.0 $\pm$ 0.0 (0)
Sweetgum	0.0 $\pm$ 0.0 (0)	0.0 $\pm$ 0.0 (0)	...	0.0 $\pm$ 0.0 (0)	...	0.0 $\pm$ 0.0 (0)	<i>3.0 $\pm$ 0.3 (82)</i>	<i>3.5 $\pm$ 0.3 (79)</i>
Olive	0.0 $\pm$ 0.0 (20)	0.0 $\pm$ 0.0 (0)	<i>2.5 $\pm$ 0.3 (86)</i>	0.0 $\pm$ 0.0 (0)	0.0 $\pm$ 0.0 (0)	0.0 $\pm$ 0.0 (0)	0.0 $\pm$ 0.0 (17)	...

^a Successful infections are shown in bold, with bold and italics indicating homologous inoculations (inoculation of a strain into its original host). Data are the mean of leaf symptoms (scale from 0 to 5; see text) across replicates plus the ELISA percent stem infection scores resulting from tests on all 7-cm sections of the stem above the inoculation point of a single plant; na = not applicable. Results shown are from week 20 for each year tested.

^b ELISA signal was weak.

^c Symptoms and ELISA were scored at week 10 (not tested in week 20).

^d No further spread was found in an ELISA test at week 60.

OLV0029 (ST7), USA, olive, 2003

Nunney et al., 2019

Genomic and phenotypical features of *Xf* subsp. *multiplex* strain ESVL (ST6)

RESOURCE ANNOUNCEMENT

*e-Xtra**

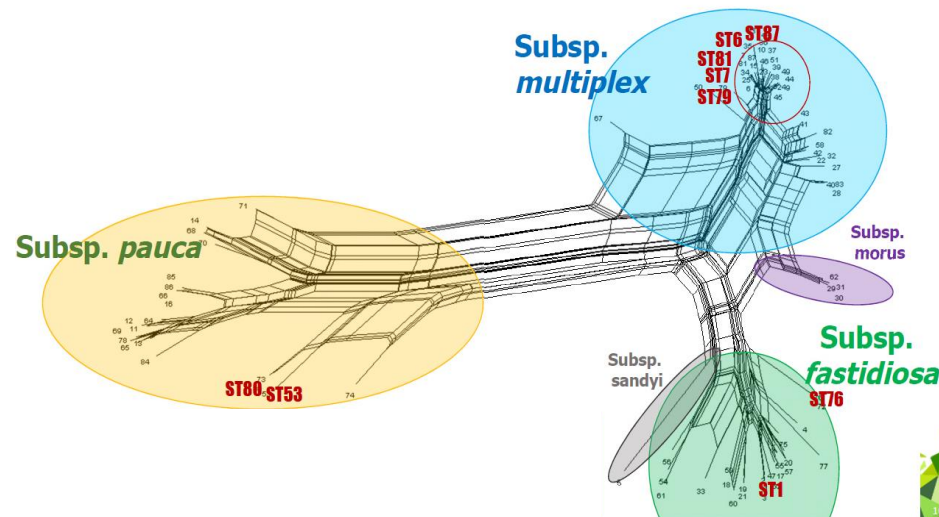
Draft Genome Resources of Two Strains (“ESVL” and “IVIA5901”) of *Xylella fastidiosa* Associated with Almond Leaf Scorch Disease in Alicante, Spain

Table 1. Summary statistics of the genome assemblies of *Xylella fastidiosa* strains IVIA5901 and ESVL^a

Strain	Number of supercontigs (≥50,000 bp)	Number of contigs	Number of contigs plasmid related	Total length (bp)	Genome coverage	N50 (bp)	N75 (bp)	Max (bp)	Min (bp)	GC content (%)	Predicted genes
ESVL	18	133	2	2,554,495	110x	103,116	67,809	335,582	207	51.8	2,515
IVIA5901	16	141	–	2,493,558	309x	116,584	67,790	335,598	218	51.8	2,464

“...two Spanish isolates (**ESVL and IVIA5901**) of *Xf* subsp. *multiplex* have lower motility, less capacity of aggregation, make less biofilm, and cause lower disease severity when compared with the US isolates (Alma-Em3 and BB08-1) in virulence assays conducted in the greenhouse using tobacco plants cvs. SR1 and Xanthi.

...ESVL exhibits stronger attachment to substrate than IVIA5901, but no differences in biofilm formation, cell–cell aggregation or twitching motility were found between them” (Román-Écija M et al., Book_of_abstracts Ajaccio EFSA 2019-1-108-69)



multiplex

ST6



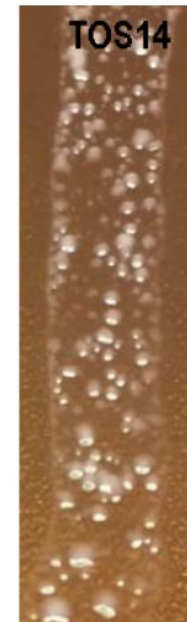
TOS4



TOS5



TOS14



pauca
(ST53)

De donno



Colonies on BCYE after 2 weeks. Courtesy M. Saponari (CNR).

Table 1. Comparative analyses of the growth rate, cell-surface attachment, and cell-to-cell aggregation yielded for *Xylella fastidiosa* subsp. *multiplex* strains ESVL, TOS4, TOS5, and TOS14^w

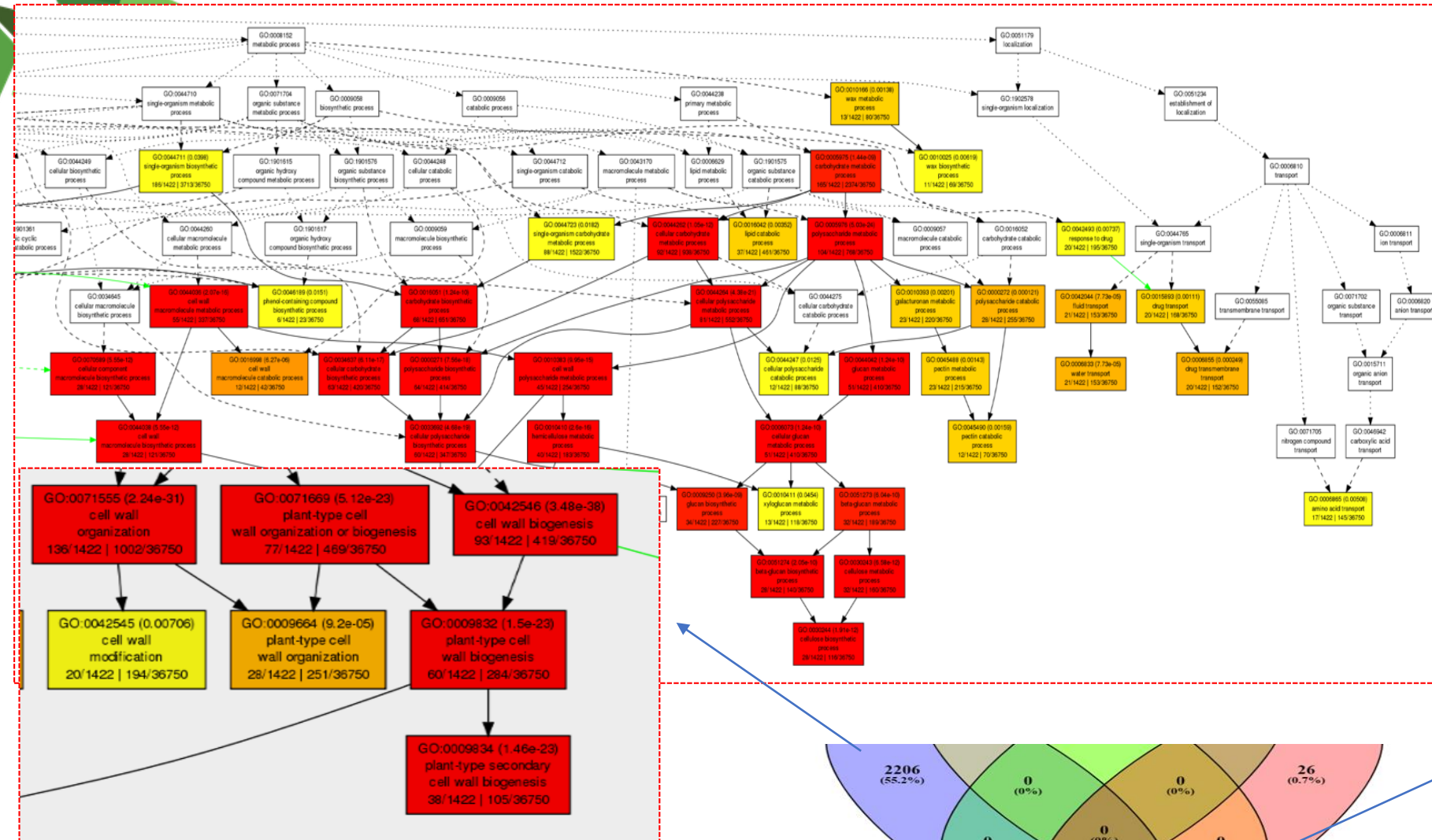
Strains	Growth rate (OD ₆₀₀ day ⁻¹) (mean ± SD) ^x	Cell-surface attachment (OD ₆₀₀) (mean ± SD) ^y	Cell-to-cell aggregation (OD ₆₀₀ min ⁻¹) (mean ± SD) ^z
ESVL	0.67 ± 0.21 a	0.15 ± 0.02 a	0.001 ± 0.00 a
TOS4	0.49 ± 0.13 a	0.18 ± 0.01 a	0.229 ± 0.02 b
TOS5	0.62 ± 0.12 a	0.14 ± 0.02 a	0.320 ± 0.08 b
TOS14	0.54 ± 0.15 a	0.15 ± 0.03 a	0.236 ± 0.03 b

ESVL has a lower cell-to-cell aggregation and grows in very small contigues colonies compared to others EU strains of subsp. *multiplex* (ST87) which tend to form large aggregated and clearly distinguished colonies

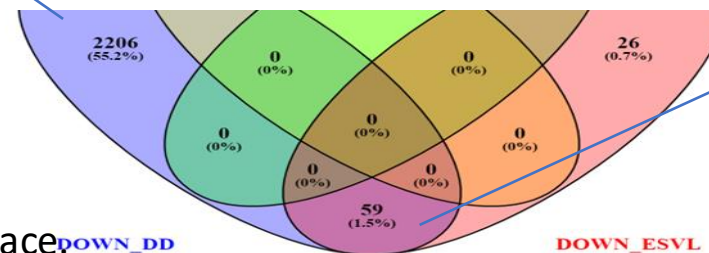
Giusy D’Attoma and Maria Saponari

In planta virulence of the pathogen relies on a fine balance between more motile bacterial forms, able to efficiently multiply and move within and between the xylem conduits, and sticky aggregative cells forming a biofilm, which are responsible for the vessels clogging and insects acquisition (Chatterjee et al., 2008, Sun et al., 2013)

GO:0008150
biological_process



GO:0010322 (2.32e-09)
regulation of
isopentenyl diphosphate biosynthetic process, methylerythritol 4-phosphate pathway
5/34 | 18/36750



DOWN ESVI