



Evaluation of field treatments to reduce the impact of *Xylella* *fastidiosa* infections in olive trees

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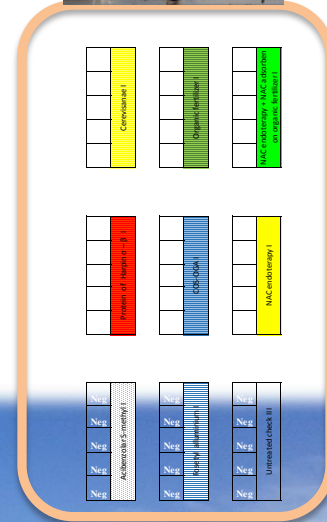
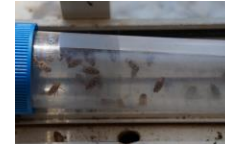
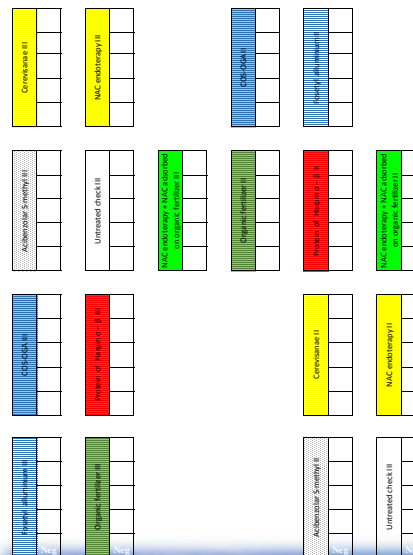
Field location and trials setup

TRIAL:	A	B	C	D
FIELD LOCATION:	Gallipoli (Le)	Nociglia (Le)	Taurisano (Le)	Ugento (Le)
CULTIVAR:	Cellina di Nardò	Cellina di Nardò	Carolea; Gioconda; Pesciolen; Leccino; Nocellara.	Carolea
PLANTING DATA:	2015	2005	2000	1986
TRAINING SYSTEM:	Open centre tree			
DISTANCE BETWEEN ROWS (m):	5 x 5	6 x 6	5 x 5	7 x 7
TRIAL DESIGN:	Randomized block	Randomized block	Completely randomized block	Randomized block
REPLICATIONS:	3	4	6	4
N. PLANTS/TREATMENT:	15	24	52	32

finding answers to a global problem

Trial A

FIELD LOCATION:	Gallipoli (Le)
CULTIVAR:	Cellina di Nardò
PLANTING DATA:	2015
TRAINING SYSTEM:	Open centre tree
DISTANCE BETWEEN ROWS (m):	5 x 5
TRIAL DESIGN:	Randomized block
REPLICATIONS:	3
N. PLANTS/TREATMENT:	15



Infected ancient olive orchard




























FIELD LOCATION:	Nociglia (Le)
CULTIVAR:	Cellina di Nardò
PLANTING DATA:	2005
TRAINING SYSTEM:	Open centre tree
DISTANCE BETWEEN ROWS (m):	6 x 6
TRIAL DESIGN:	Randomized block
REPLICATIONS:	4
N. PLANTS/TREATMENT:	24












Trial b



































Trial B: February 2016 (at the beginning of the trial)












Fosetyl Aluminium IV		Cerevisanae IV				Protein of Harpin IV	Organic fertilizer IV
  		  				 	  















	Untreated check IV		COS-OGA IV	NAC endotherapy IV		Acibenzolar S-methyl IV	NAC endotherapy adsorbed on organic fertilizer IV
			 	  		   	   














		Organic fertilizer III	NAC endotherapy adsorbed on organic fertilizer III	Cerevisanae III		NAC endotherapy III	
		  	 	  		  	










	COS-OGA III	Fosetyl Aluminium III	A. tenuissima - low dose III	Laminarin - low dose III	Protein of Harpin III		Untreated check III
	   	  			  		  

Protein of Harpin II	Acibenzolar S-methyl II	COS-OGA II	NAC endotherapy II		NAC endotherapy adsorbed on organic fertilizer II		Acibenzolar S-methyl III
   	 	  	  		  		   

		Cerevisanae II		Organic matter II	Fosetyl aluminium AI II		
		   		  	   		

NAC endotherapy I			Fosetyl Aluminium I	NAC endotherapy adsorbed on organic fertilizer I	Untreated check II		
   			  	  	   		

Organic fertilizer I	Acibenzolar S-methyl I					Protein of Harpin I
   	    					   

Untreated check I			Cerevisanae I		COS-OGA I
  			 		   



Trial C

FIELD LOCATION:	Taurisano (Le)
CULTIVAR:	Carolea; Gioconda; Pesciolen; Leccino; Nocellara.
PLANTING DATA:	2000
TRAINING SYSTEM:	Open centre tree
DISTANCE BETWEEN ROWS (m):	5 x 5
TRIAL DESIGN:	Completely randomized block
N. PLANTS/TREATMENT:	52

Infected ancient olive orchard

Trial field



Foreshorening of trial field

The farmer implemented the best agricultural practice, with:

- adequate control of the weeds;
- frequently pruning, both during the winter in the season by removing desiccation shoots;
- Insecticides applications against mainly pests of olive (Dacus oleae; Prays oleae; Prays oleae);



Trial D

TRIAL:	D
FIELD LOCATION:	Ugento (Le)
CULTIVAR:	Carolea
PLANTING DATA:	1986
TRAINING SYSTEM:	
DISTANCE BETWEEN ROWS:	7 x 7
TRIAL DESIGN:	Randomized block
REPLICATIONS:	4
N. PLANTS/TREATMENT:	32





TESTED PRODUCTS

ELICITORS OF PLANTS DEFENSE

Product name or code	Active substance (a.s.)	Content a.s. (% or g/L) and formulation	Applied volume (mL/ha)	Formulate rate (g or mL/hL)	Mode of application	Trial			
						A	B	C	D
ALD 1901	Cerevisanae	(100%) WP	1000 L/ha	75 g/hL	Spray	X	X		X
Aliette	Fosetyl alluminium	(80%) WG	1000 L/ha	300 g/hL	Spray	X	X	X	X
HP400	Protein of Harpin $\sigma - \beta$	(1%) PB	1000 L/ha	20 g/hL	Spray	X	X	X	X
Ibisco	COS-OGA	(12 g/L) L	1000 L/ha	500 mL/hL	Spray	X	X	X	
Bion	Acibenzolar S-Methyl	(50%) WG	1000 L/ha	20 g/hL	Spray	X	X	X	X

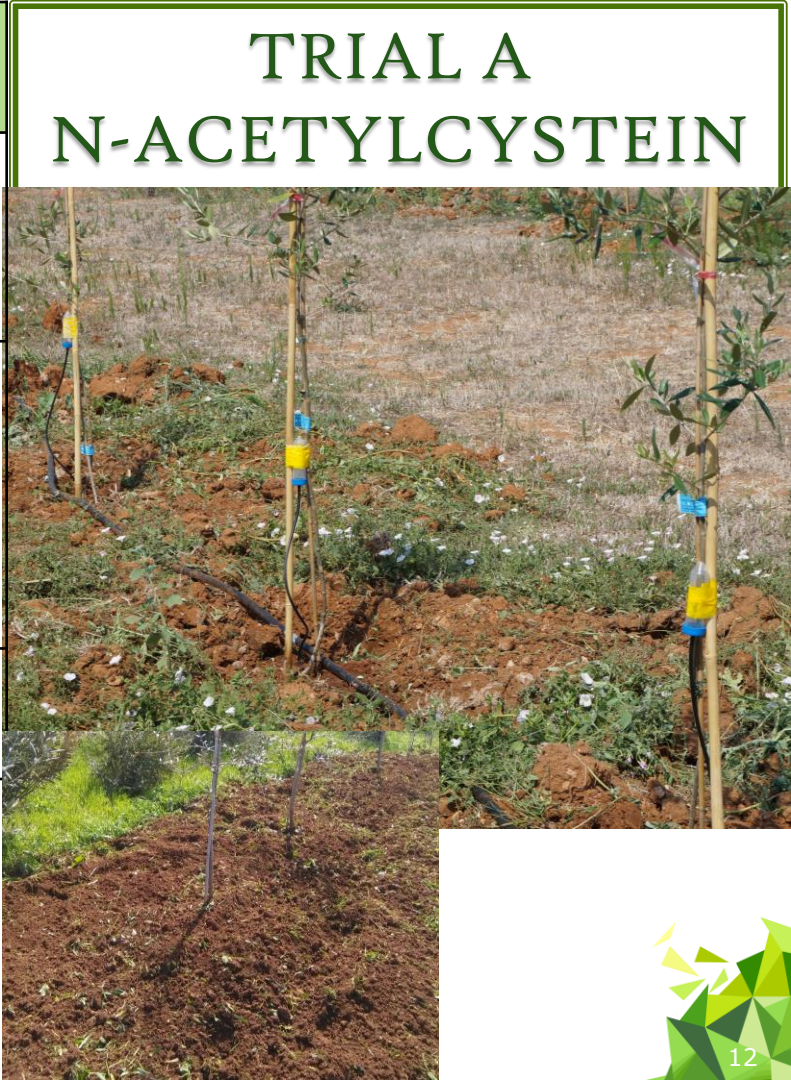
- Intervals applications: 15–20 days;
- Starting on the end of the winter (new shoots emergence) until July, interruption on august, 2 applications in September/October;
- Starting trials:
 - Trial A: 2015 until 2017;
 - Trial B: 2016 to the end of february 2017;
 - Trial C: 2015 until 2017;
 - Trial D: 2016 until 2017;



N-ACETYL CYSTEINE

Product name or code	Active substance (a.s.)	Content a.s. (% or g/L) and formulation	Mode of applications	Trial			
				A	B	C	D
NAC	N-acetylcystein	(22.5 g/L) L	Endotherapy	X	X		
NAC + [NAC + Organic fertilizer]	N-acetylcystein + [N-acetylcystein + organic fertilizer]	(45 g/L) L + [organic N (4%); organic C (41%); organic mater (70%)] Pellet	Endotherapy and incorporated in the soil, adsorbed on organic fertilizer	X	X		
Pollina	Organic fertilizer	[organic N (4%); organic C (41%); organic mater (70%)] Pellet	Incorporated in the soil	X	X		

Active substance (a.s.)	Applied volume (mL/plant)	Formulate rate (g or mL/plant)	Mode of application
N-acetylcystein	50 mL/plant (2015-2016) 100 mL/plant (2017)	1.125 g/plant (2015-2016) 2.25 g/plant (2017)	Endotherapy
N-acetylcystein + [N-acetylcystein + organic fertilizer]	50 mL/plant (2015-2016) 100 mL/plant (2017)	2.25 g/plant (2015-2016) 4.5 g/plant (2017) + [4 g/plant + 400 g/plant]	Endotherapy and incorporate in the soil
Organic fertilizer	-	400 g/plant	Incorporate in the soil



TRIAL A

N-ACETYLCYSTEIN

- Endotherapy: single application in summer 2015; two applications in spring 2016 and 2017;
- NAC adsorbed on organic fertilizer and organic fertilizer alone: starting application in 2016, seven application both in 2016 and 2017, started in the spring to autumn with 15–20 days intervals;



TRIAL B: N-ACETYL CYSTEINE

Active substance (a.s.)	Applied volume (mL/plant)	Formulate rate (g or mL/plant)	Mode of application
N-acetylcystein	500 mL/plant	11.25 g/plant	Endoterapy
N-acetylcystein + [N-acetylcystein + organic fertilizer]	500 mL/plant	22.5 g/plant + [4 g/plant + 4000 g/plant]	Endoterapy and incorporate in the soil
Organic fertilizer	-	4000 g/plant	Incorporate in the soil

- Endotherapy: in 2016, four applications started in the spring with 20
- 30 days intervals;



TRIAL B: N-ACETYL CYSTEINE

Active substance (a.s.)	Applied volume (mL/plant)	Formulate rate (g or mL/plant)	Mode of application
N-acetylcystein	500 mL/plant	11.25 g/plant	Endoterapy
N-acetylcystein + [N-acetylcystein + organic fertilizer]	500 mL/plant	22.5 g/plant + [4 g/plant + 4000 g/plant]	Endoterapy and incorporate in the soil
Organic fertilizer	-	4000 g/plant	Incorporate in the soil

- In 2016: NAC adsorbed on organic fertilizer and organic fertilizer alone: eight applications from spring to autumn with 15–20 gg intervals;



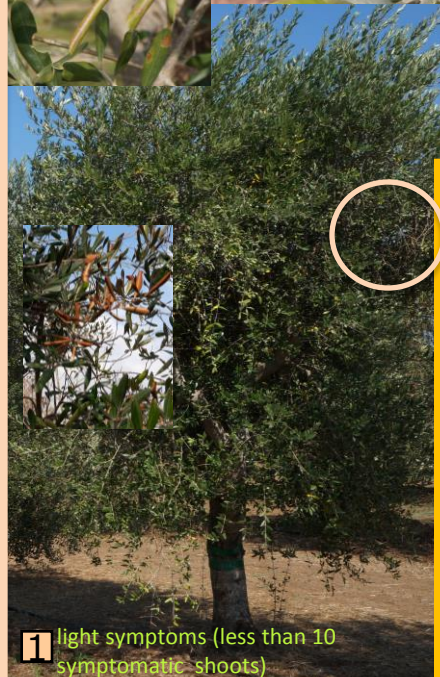


0 symptomless

Empyrical scale



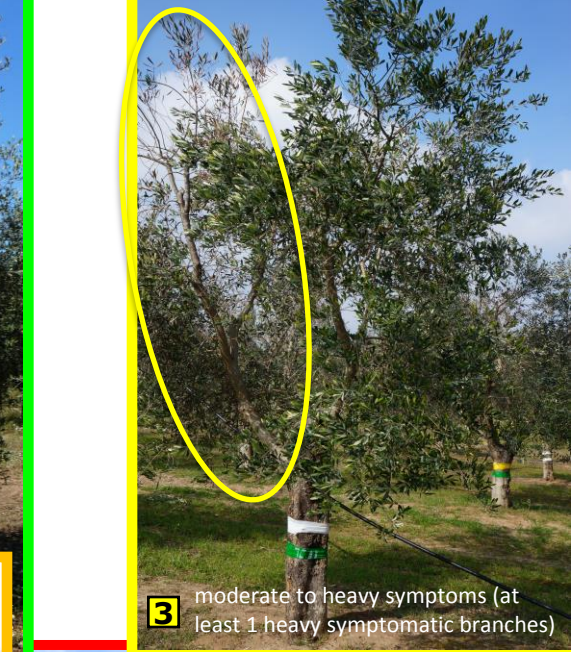
1 light symptoms (less than 10 symptomatic shoots)



2 light to moderate symptoms (more than 10 symptomatic shoots)



4 heavy symptoms or branches (at least 2 symptomatic branches)

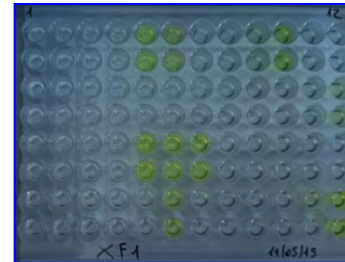
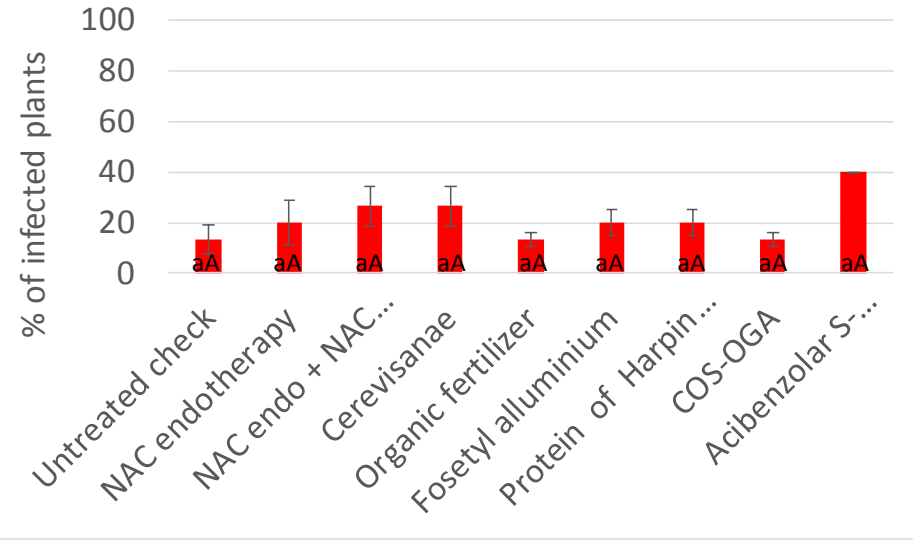
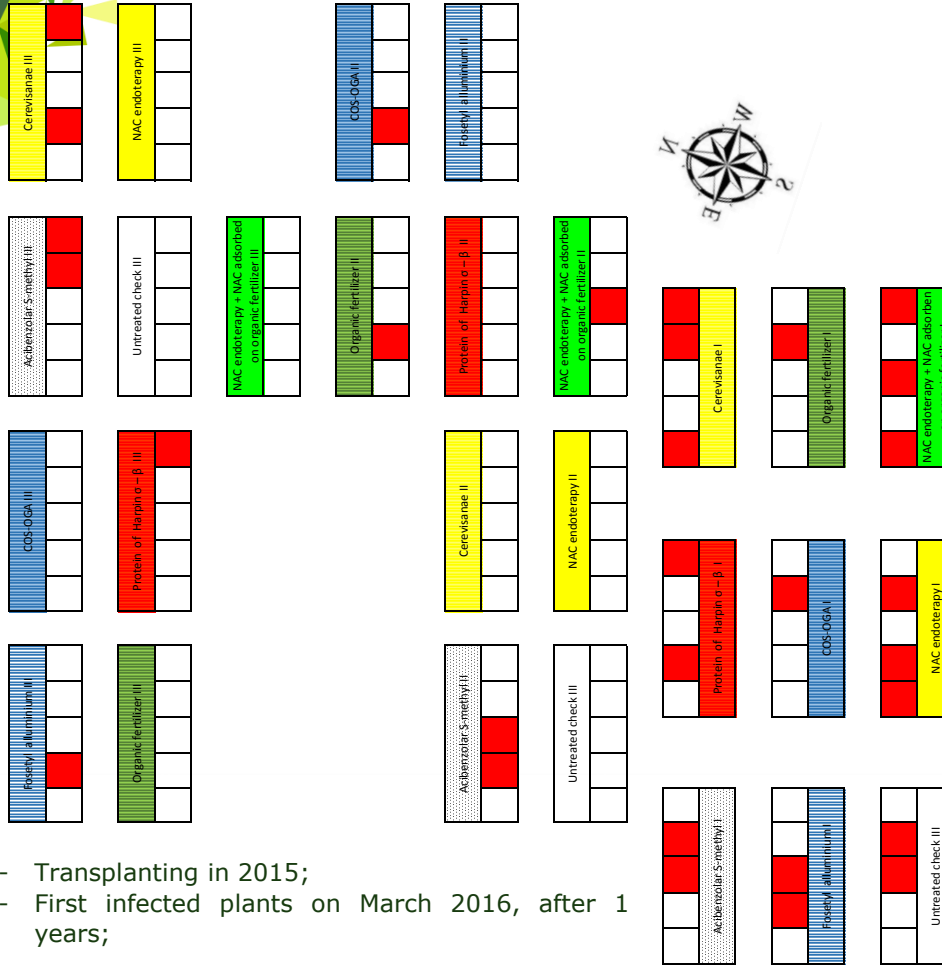


3 moderate to heavy symptoms (at least 1 heavy symptomatic branches)



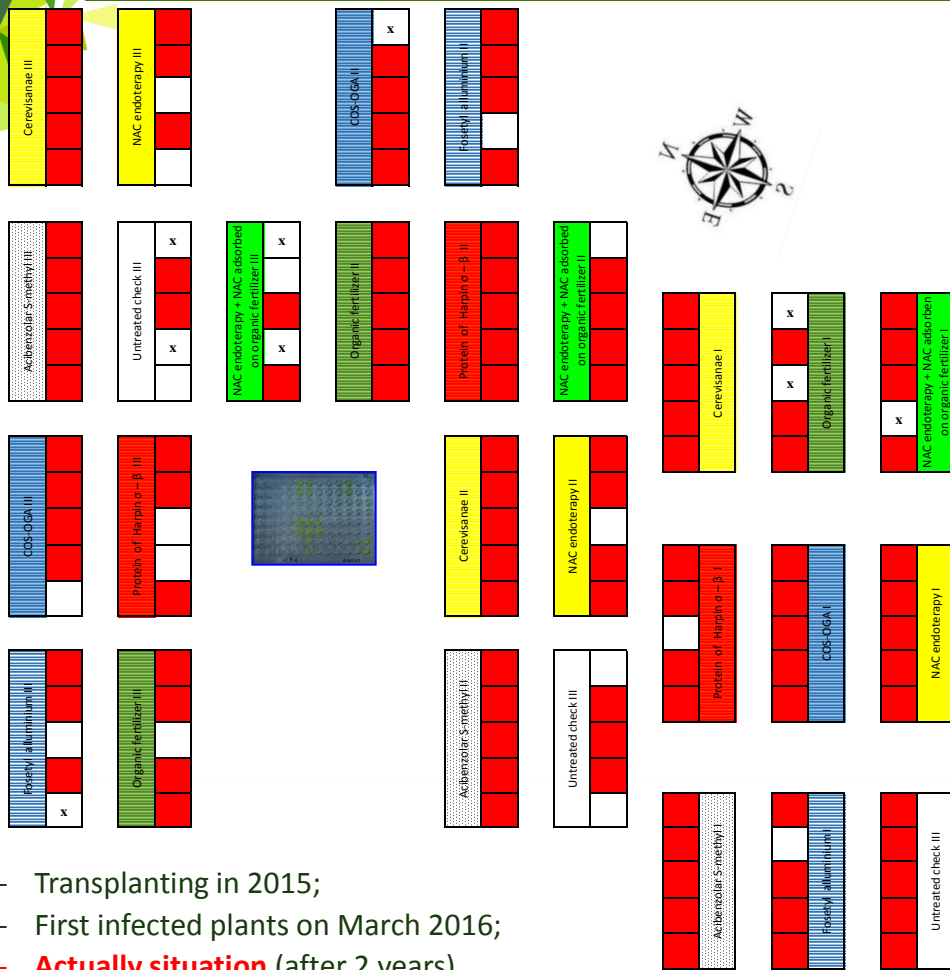
5 heavy symptoms on the whole tree

Trial A: March 2016

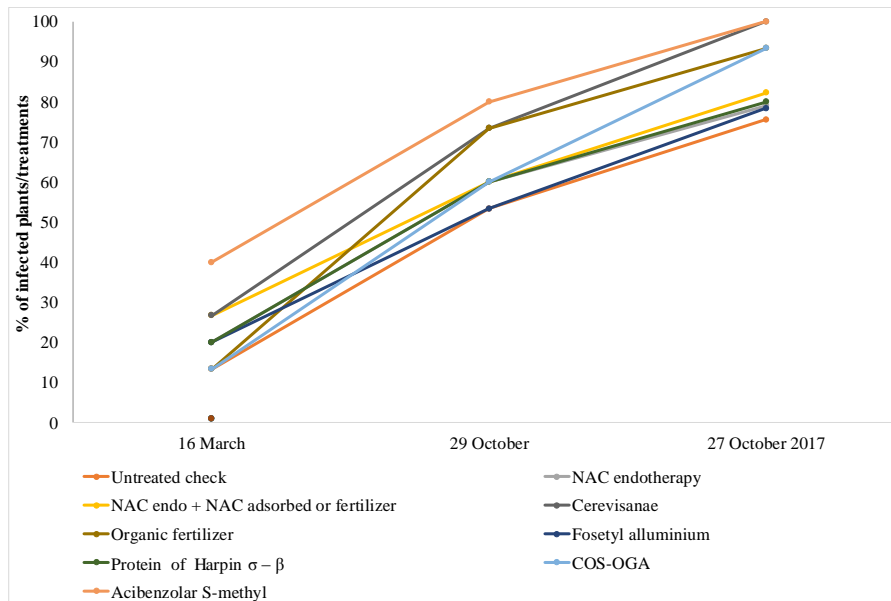


- Transplanting in 2015;
- First infected plants on March 2016, after 1 years;

Trial A: September 2017

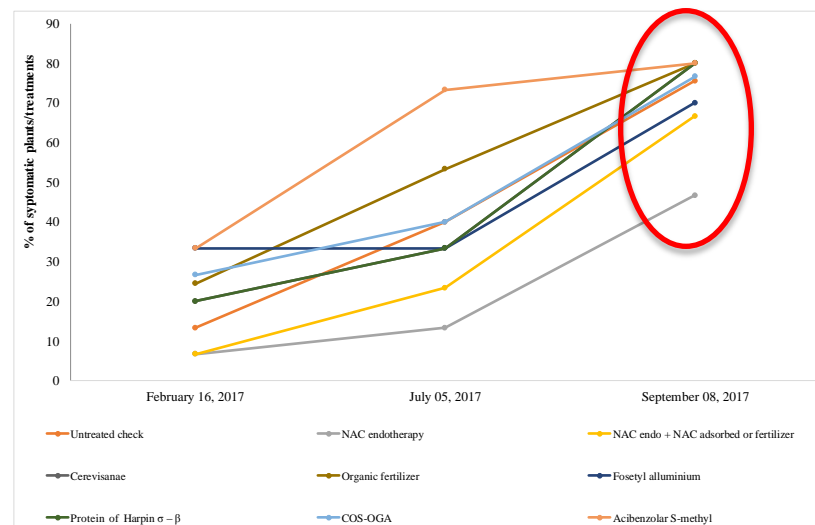
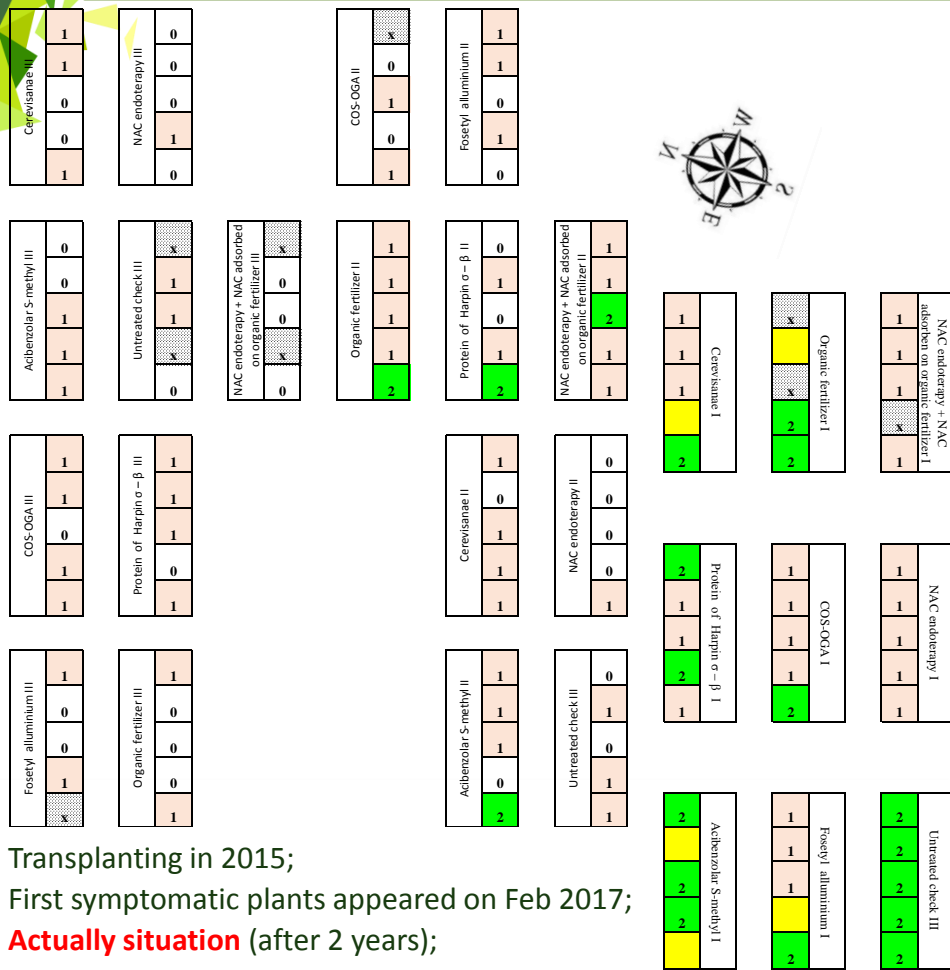


Active substance (a.s.)	October 2017	
Untreated check	75.6	b A
NAC endotherapy	79.0	b A
NAC endo + NAC adsorbed or fertilizer	82.2	ab A
Cerevisanae	100.0	a A
Organic fertilizer	93.3	ab A
Fosetyl aluminium	78.3	ab A
Protein of Harpin $\sigma - \beta$	80.0	ab A
COS-OGA	93.3	ab A
Acibenzolar S-methyl	100.0	a A



- Transplanting in 2015;
- First infected plants on March 2016;
- **Actually situation** (after 2 years)

Trial A: September 2017: prevalence (%)



Empirical scale: 0

1

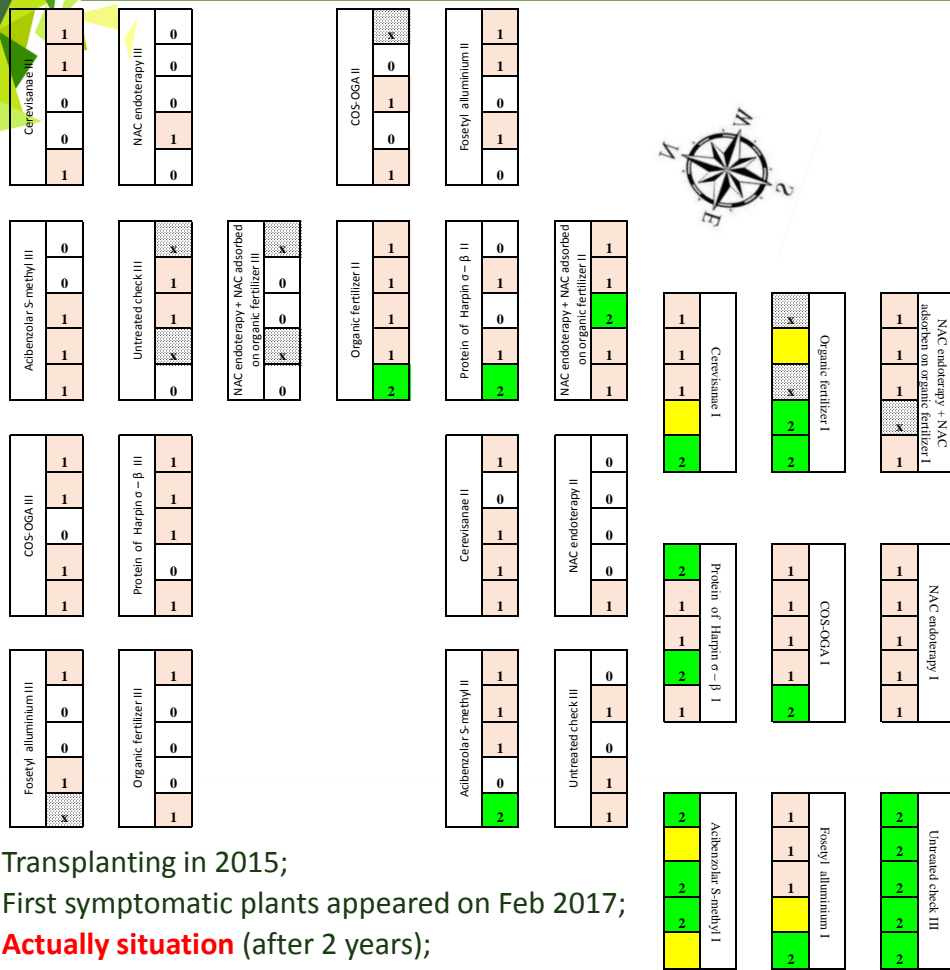
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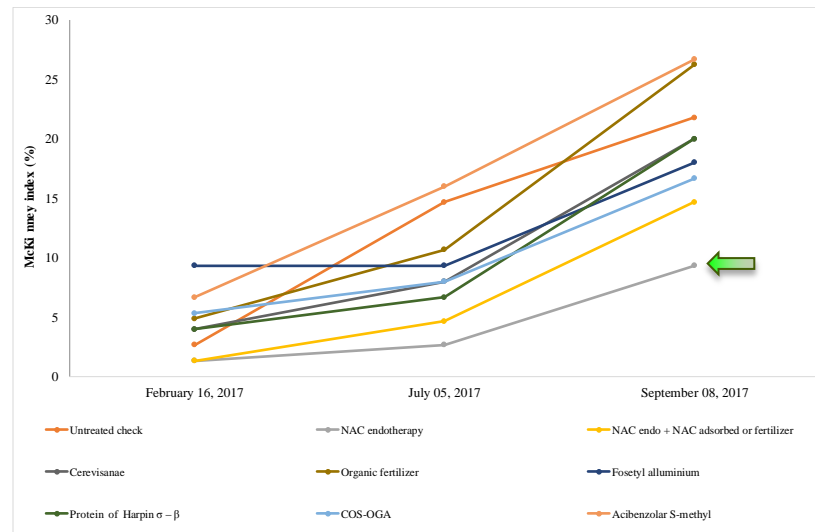
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5

Trial A: September 2017: McK (



McKinney Index (%)	
Active substance (a.s.)	September 2017
Untreated check	21.7 a A
NAC endotherapy	9.3 b A
NAC endo + NAC adsorbed or fertilizer	14.7 ab A
Cerevisanae	20 a A
Organic fertilizer	26.2 a A
Fosetyl aluminium	18 ab A
Protein of Harpin $\sigma - \beta$	20 a A
COS-OGA	16.7 ab A
Acibenzolar S-methyl	26.7 a A

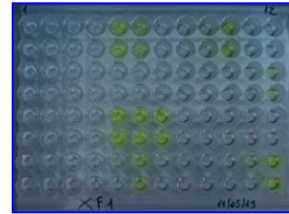
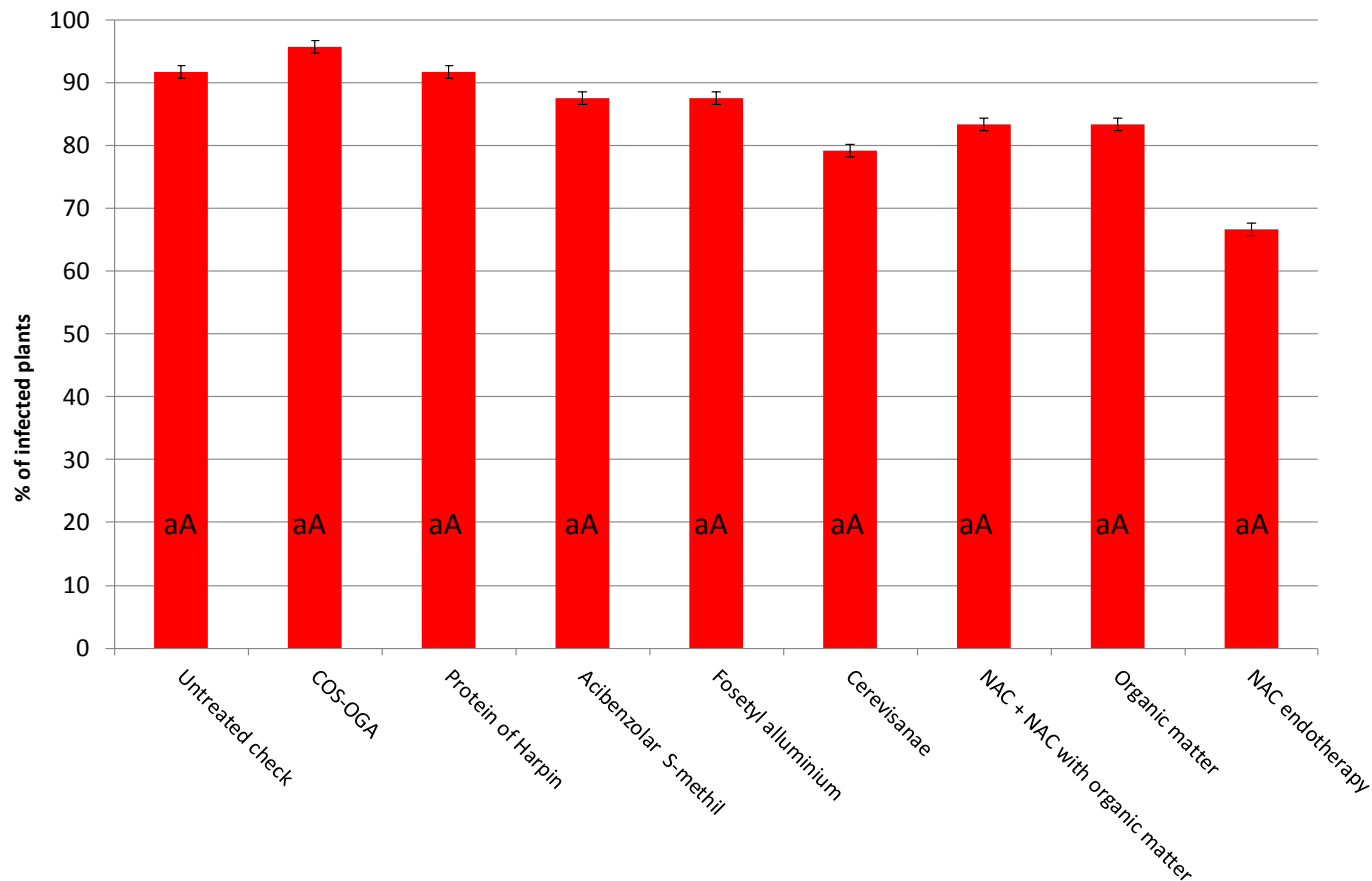


Empirical scale: 0 1 2 3 4 5

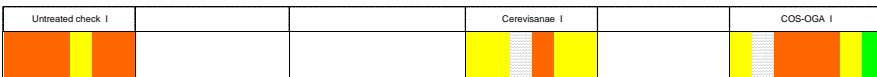
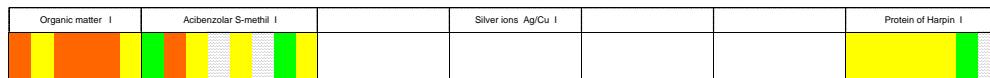
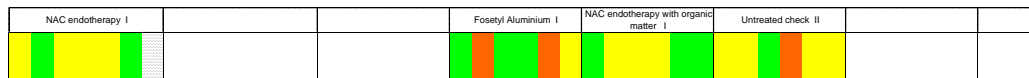
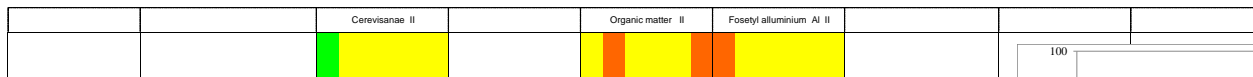
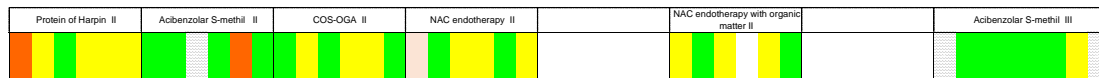
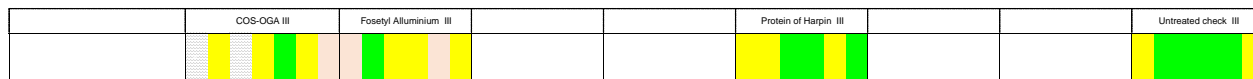
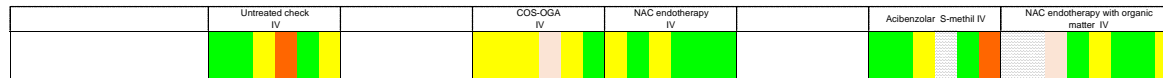
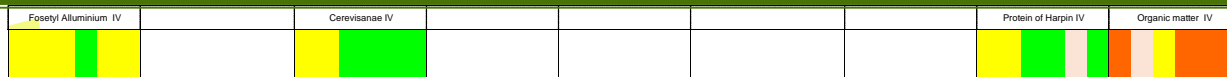
Trial A: EVOLUTION IN the time



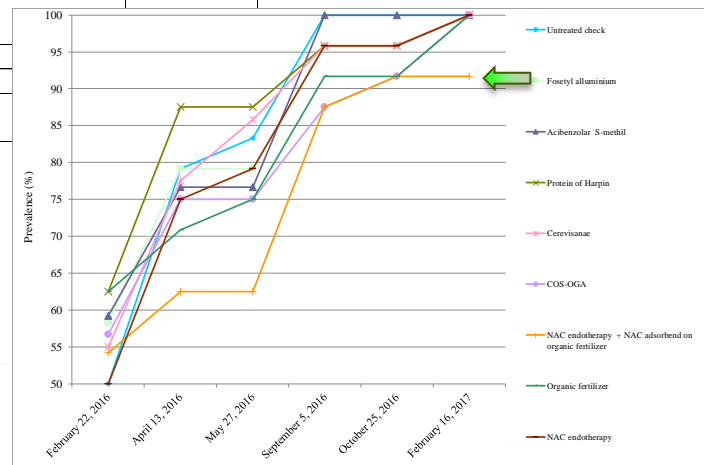
Trial B: november 2016 (infected plants after 10 months from the start the trial)



Trial B: February 2017 (after 1 years from the start the trial)

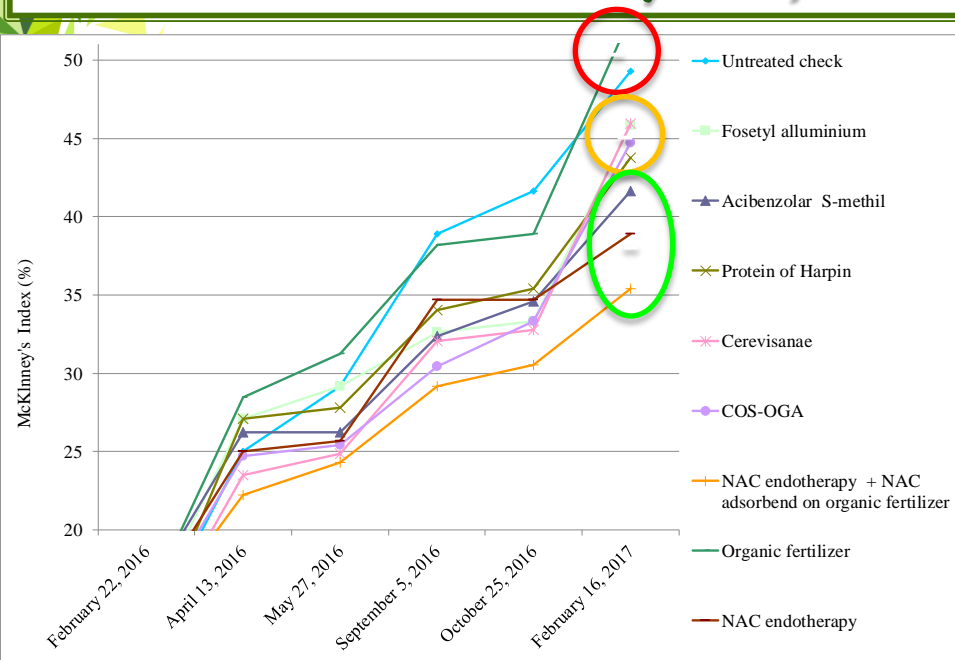


Treatment	Prevalence (%)		
	16/02/2017		
Untreated check	100	a	A
Fosetyl aluminium	100	a	A
Acibenzolar S-methyl	100	a	A
Protein of Harpin	100	a	A
Cerevisanae	100	a	A
COS-OGA	100	a	A
NAC endotherapy + NAC adsorbed on organic fertilizer	91,7	b	B
Organic fertilizer	100	a	A
NAC endotherapy	100	a	A



Empyrcal scale: 0 1 2 3 4 5

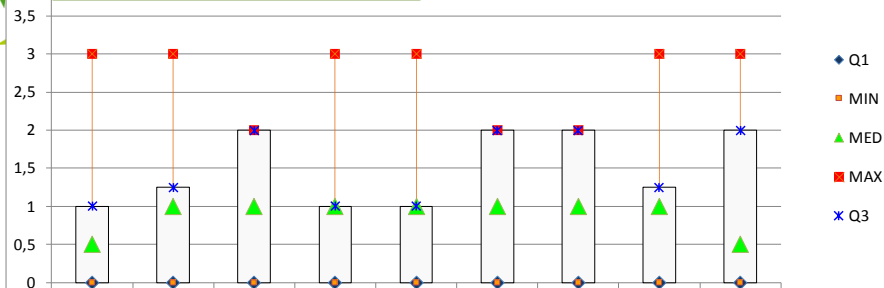
Trial B: February 2017



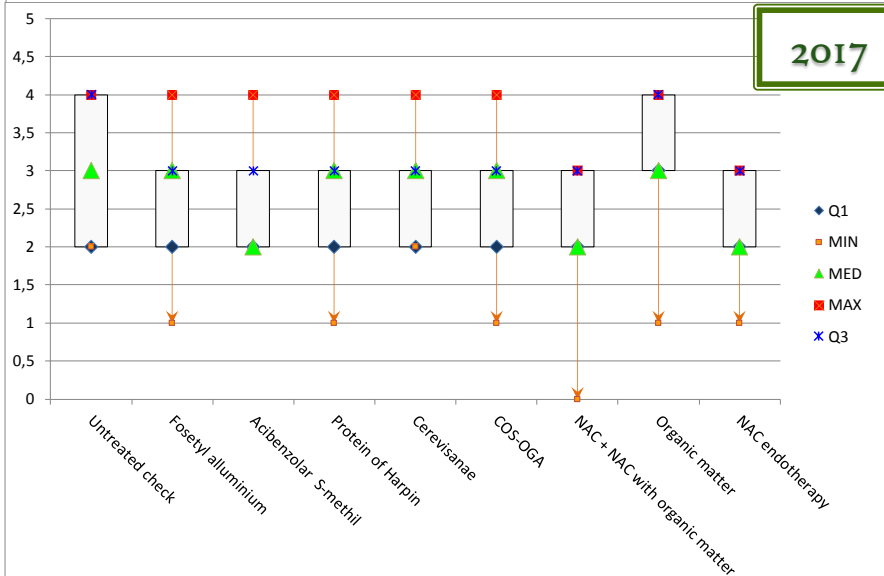
Treatment	Mckinney index (%) 16/02/2017		
Untreated check	49,3	ab	AB
Fosetyl alluminium	45,8	bc	ABC
Acibenzolar S-methyl	41,7	cd	BCD
Protein of Harpin	43,8	bc	ABCD
Cerevisanae	46,0	abc	ABC
COS-OGA	44,7	bc	ABCD
NAC endotherapy + NAC adsorbed on organic fertilizer	35,4	d	D
Organic fertilizer	52,8	a	A
NAC endotherapy	38,9	cd	CD

Trial B

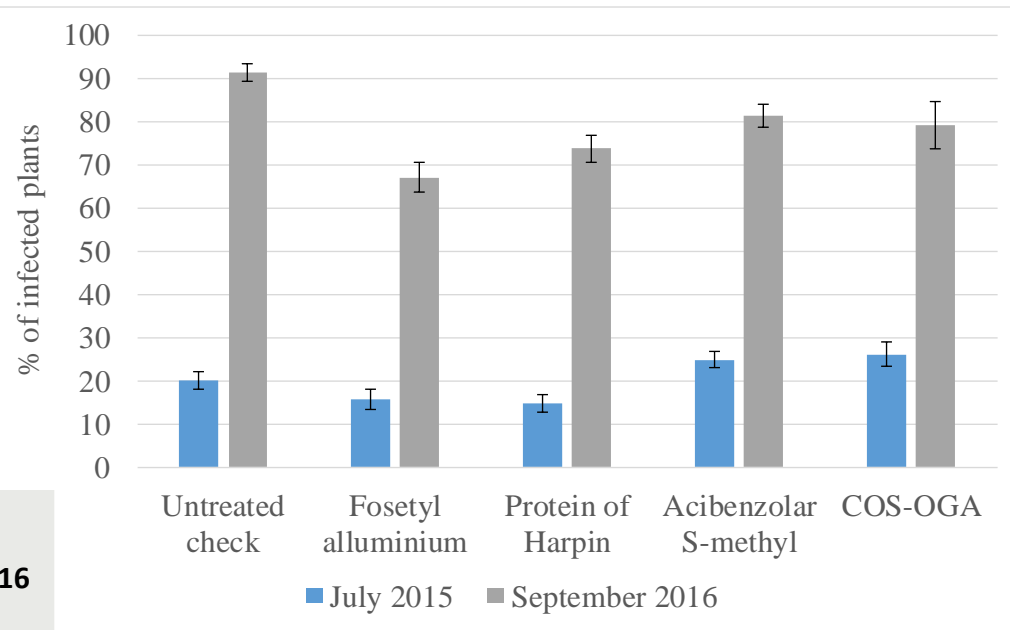
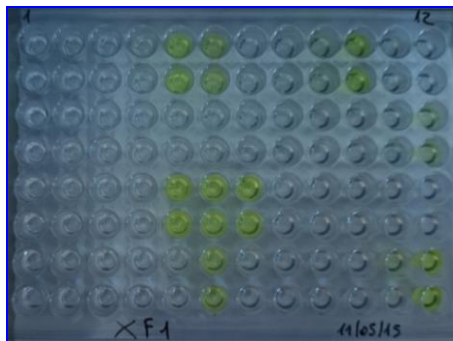
2016



2017

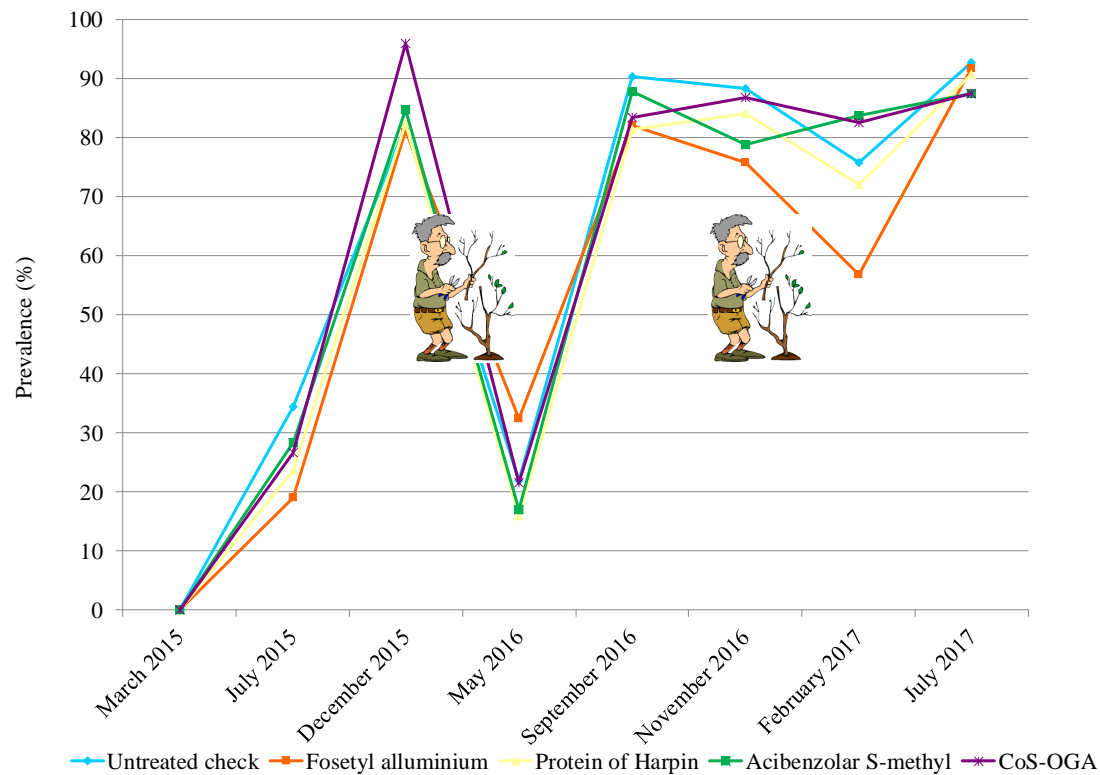


Trial C



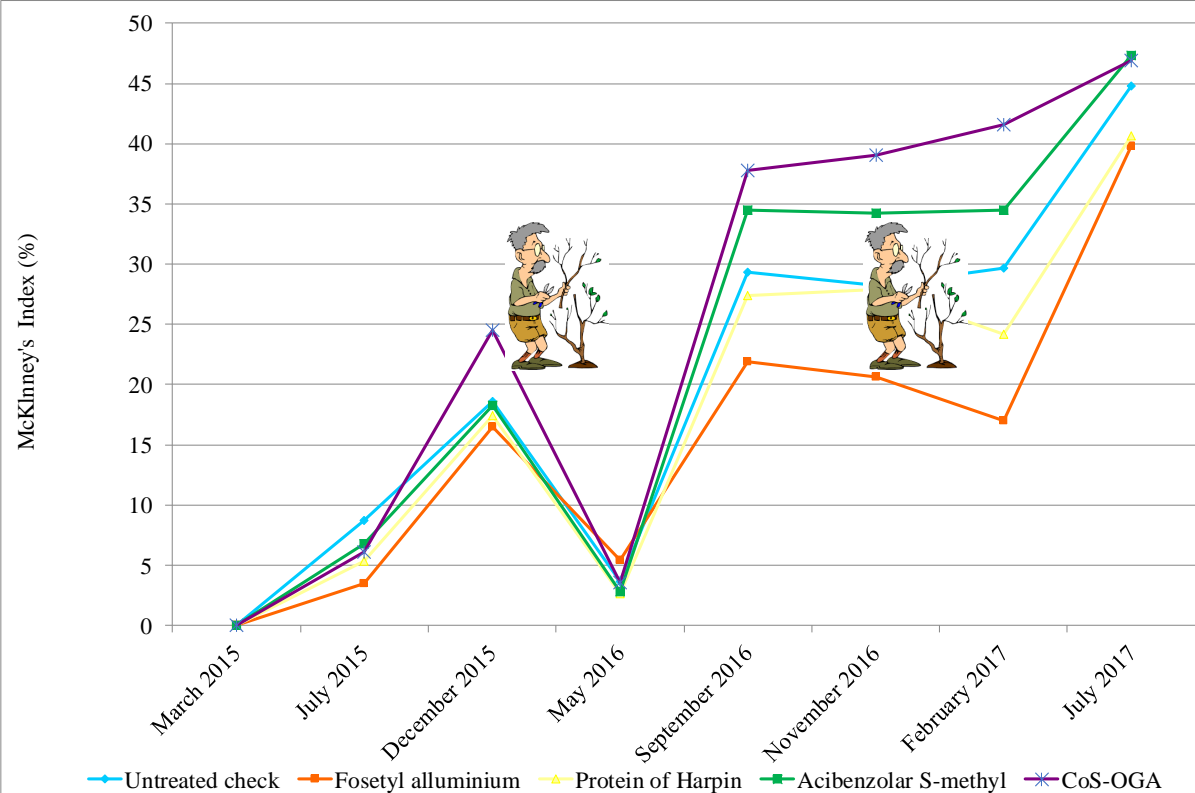
Active substance (a.s.)	July 2015		September 2016	
Untreated check	20,1	a A	91,3	a A
Fosetyl alluminium	15,7	a A	67,1	b A
Protein of Harpin	14,9	a A	73,7	ab A
Acibenzolar S-methyl	24,9	a A	81,3	ab A
COS-OGA	26,2	a A	79,2	ab A

Trial C



Thesis	Prevalence (%)															
	March 2015		July 2015		December 2015		May 2016		September 2016		November 2016		February 2017		July 2017	
Untreated check	0	a A	34,4	a A	81,9	b A	21,9	a A	90,3	a A	88,2	ab A	75,7	ab A	92,7	a A
Fosetyl alluminium	0	a A	19,0	a A	81,1	b A	32,4	a A	82,1	a A	75,8	b A	56,8	b A	91,7	a A
Protein of Harpin	0	a A	23,6	a A	82,4	b A	15,9	a A	81,3	a A	84,0	ab A	72,0	ab A	90,6	a A
Acibenzolar S-methyl	0	a A	28,3	a A	84,8	b A	16,9	a A	87,8	a A	78,8	ab A	83,7	a A	87,5	a A
CoS-OGA	0	a A	26,6	a A	95,8	a A	21,5	a A	83,3	a A	86,7	a A	82,5	a A	87,5	a A

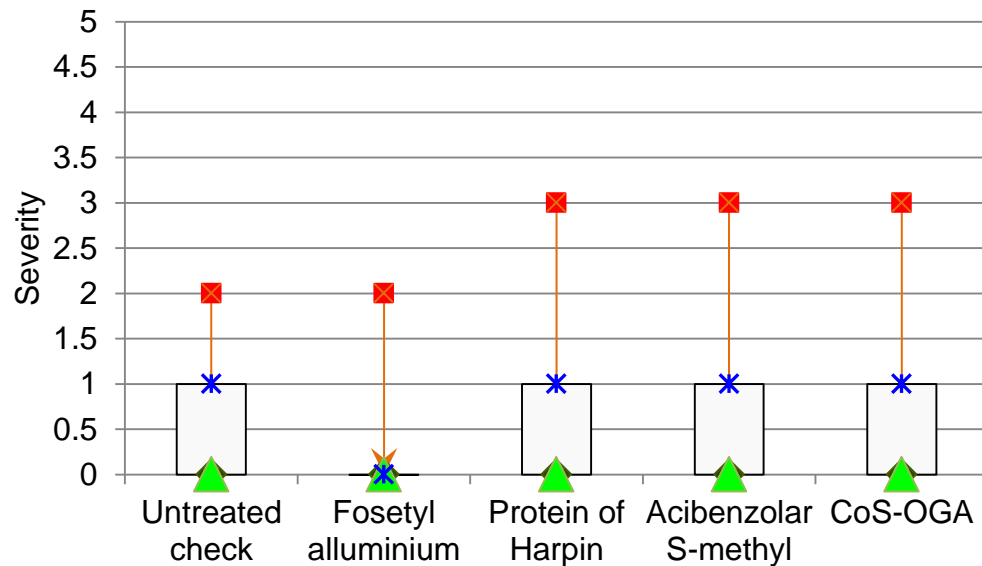
Trial C



Tesi	McKinney Index (%)															
	March 2015		July 2015		December 2015		May 2016		September 2016		November 2016		February 2017		July 2017	
Untreated check	0	a A	8,7	a A	18,6	ab A	3,6	a A	29,3	a A	28,3	ab AB	29,7	ab ABC	44,8	a A
Fosetyl alluminium	0	a A	3,5	a A	16,5	b A	5,4	a A	21,9	a A	20,6	b B	17,0	c C	39,8	a A
Protein of Harpin	0	a A	5,3	a A	17,4	b A	2,7	a A	27,4	a A	27,9	ab AB	24,2	bc BC	40,6	a A
Acibenzolar S-methyl	0	a A	6,8	a A	18,2	ab A	2,8	a A	34,4	a A	34,2	a AB	34,5	ab AB	47,3	a A
CoS-OGA	0	a A	6,1	a A	24,5	a A	3,6	a A	37,8	a A	39,0	a A	41,6	a A	46,9	a A

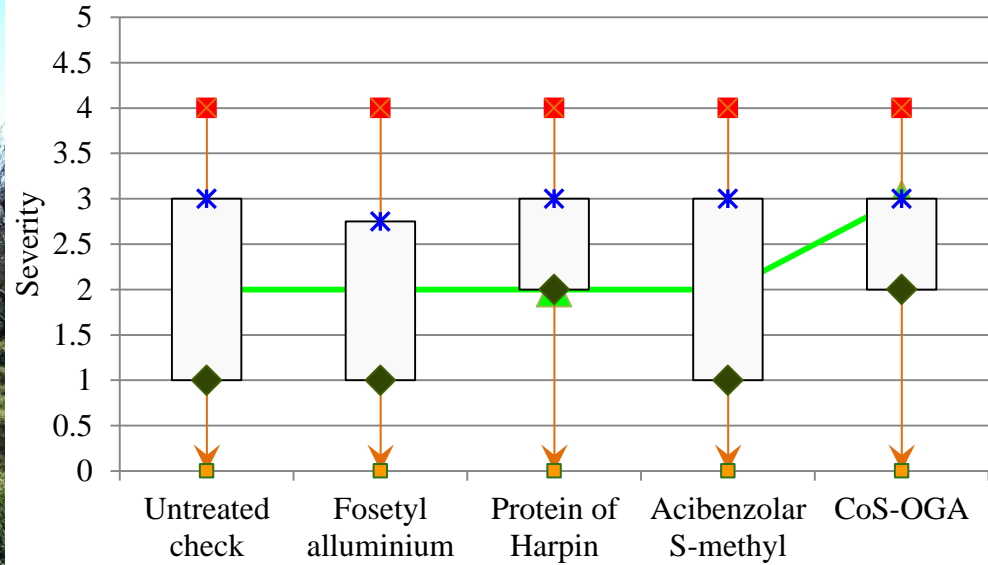
Trial C

July 2015

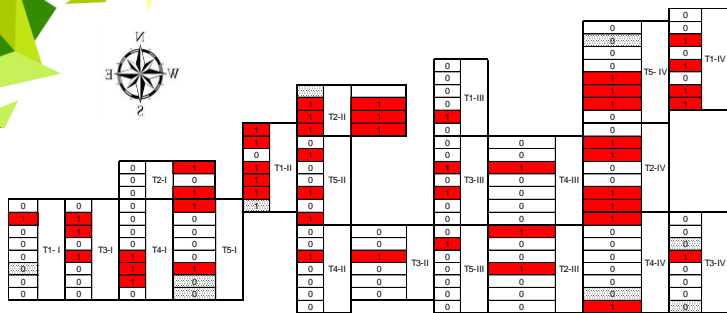


Trial C

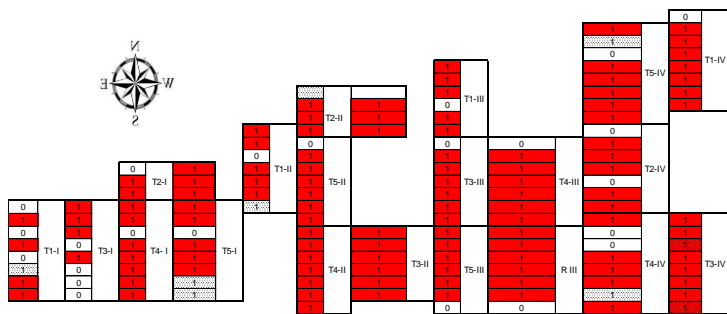
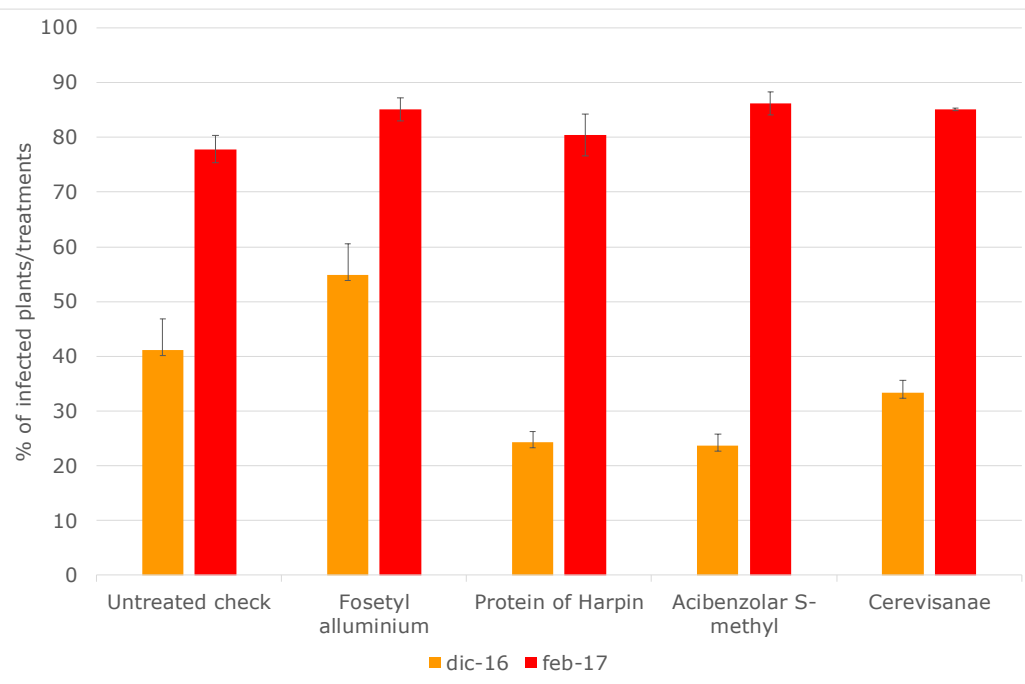
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Trial D



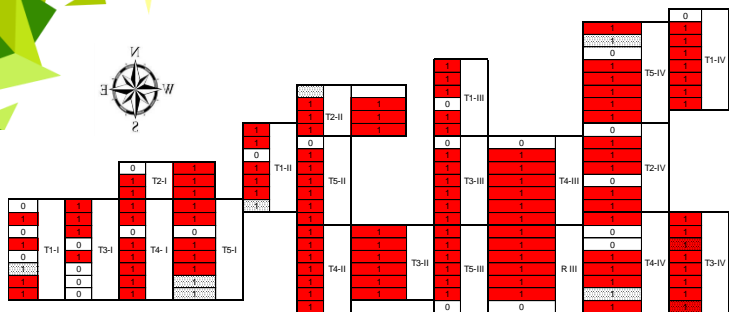
Distribution of infected plants in the field trial 1 year after the start of applications



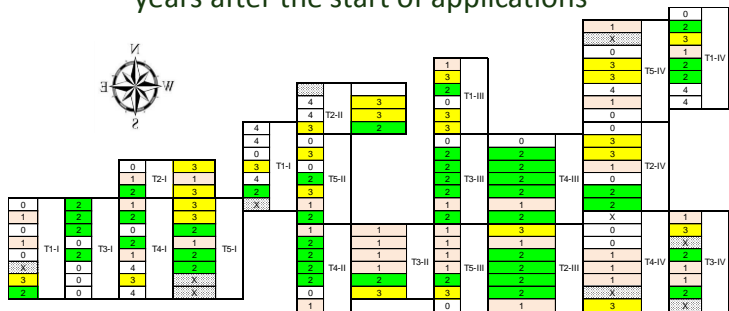
Distribution of infected plants in the field trial 2 year after the start of applications

Thesis	Dic 2016	Feb 2017
Untreated check	41.1 a A	77.8 a A
Fosetyl alluminium	54.8 a A	85.1 a A
Protein of Harpin	24.3 a A	80.4 a A
Acibenzolar S-methyl	23.7 a A	86.2 a A
Cerevisanae	33.3 a A	85.1 a A

Trial D

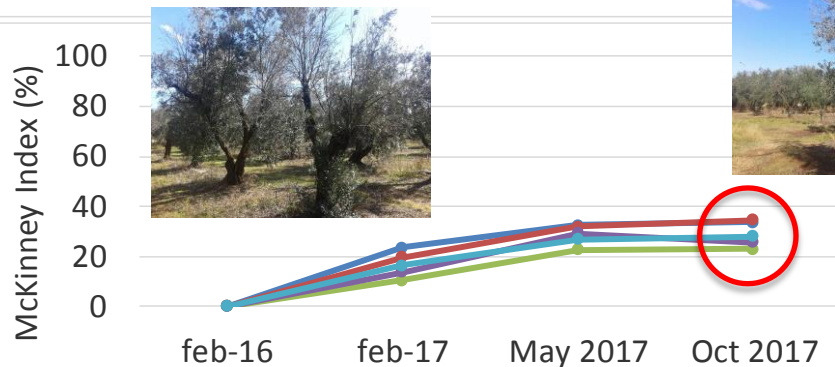
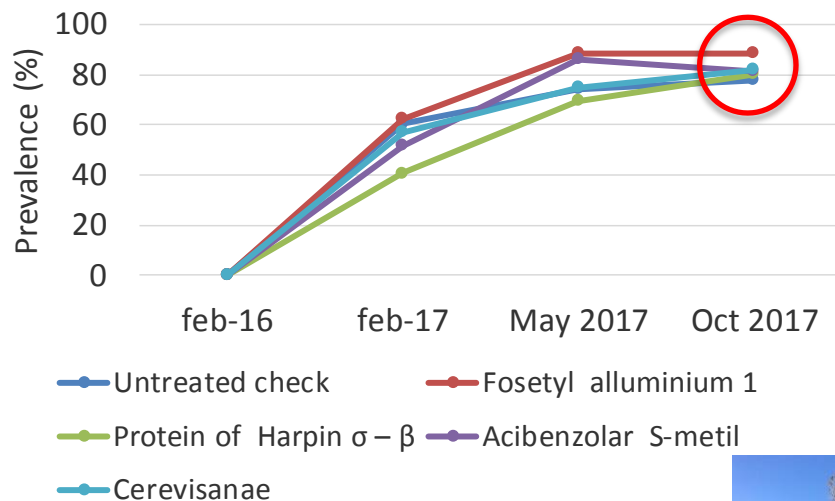


Distribution of infected plants ■ in the field trial 2 years after the start of applications




Distribution of symptomatic plants in the field trial 2 year after the start of applications

Empirical scale: 0 1 2 3 4 5



Untreated check Fosetyl aluminium 1
Protein of Harpin $\sigma - \beta$ Acibenzolar S-metil
Cerevisanae



The trial field have been set up in different olive orchard, without or with evident symptoms of *X. fastidiosa* and for different periods of times.

- resistance inductors: fosetyl aluminium, Acibenzolar S-methyl;
- Resistance inductors: COS-OGA, Protein of Harpin, Cerevisanae;
- Organic fertilizer;
- NAC;

Others experiments regarding NAC are now ongoing, in order to verify:

- others methods of application (feritrrigation), less laborious and expensive as endotherapy;
- others substrate (inert substrate) for adsorption of NAC and slowly release of the molecules;
- possibility of combination with others products (es. Sylver ions) for increasing the positive remission effects;
- different rates, time and interval of applications;

In conclusion, although, results regarding NAC was promising, actually, none of the tested products can be indicated for the efficacy containment or remission of the disease symptoms of *X. fastidiosa*.

THANKS FOR YOUR ATTENTION!!!

THANKS TO ALL MY COLLEAGUES FOR THE INTENSE AND FRUITFUL WORK THEY HAVE DONE

