

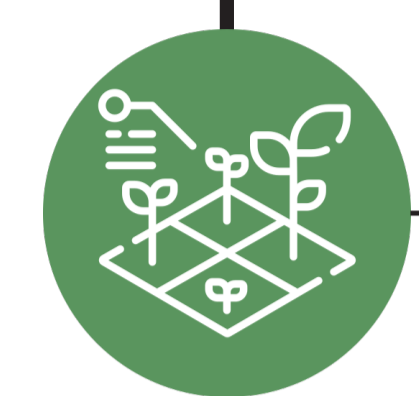
# Va.Po.Re. a project to improve the breeding of organic processing tomato

Enrico Belfanti<sup>1</sup>, Giacomo Soave<sup>1</sup>, Martina Palazzo<sup>1</sup>, Loredana Sigillo<sup>2</sup>, Katia Di Prodi<sup>3</sup>, Sara Bonora<sup>3</sup>, Stefania Delvecchio<sup>4</sup>

<sup>1</sup> TERA SEEDS srl, Gambettola (FC), Italy, <sup>2</sup> Consiglio per la ricerca in agricoltura e l'analisi dell'economia agraria - Centro di ricerca Orticoltura e Florovivaismo, Pontecagnano (SA), Italy, <sup>3</sup> CONSERVE ITALIA, San Lazzaro (BO), Italy, <sup>4</sup> RI.NOVA, Cesena (FC), Italy

Organic tomato products are highly requested by the consumers, that allow to pay extra-prize for a food free of pesticide and more environmental- oriented. Growing organic processing tomato refer to new method of taking care of soil and plants, the main difference is the exclusion of many chemical products that could reduce the incidence of pathogens. At the moment the farmers use varieties that have been constituted for traditional cultivation. The project has the goal to evaluate new genetic materials that could better perform in organic cultivation. Phytophthora infestans (Late blight - LB) and Alternaria solani (Early blight - EB) resistances are evaluate carefully by monitoring natural infection and by artificial inoculation. A new method for alternaria infection has been developed. 8 large organic tomato trials have been assessed in north of Italy to understand the agronomic performance of 30 new varieties. 4 varieties have been professional processed to get information on the quality and on the yield once processed in paste, puree, juice and dice. A new method for detecting alternaria toxin has been developed.

## 1st YEAR



### 01 Trial Fields

20 processing tomato varieties developed by Tera Seeds are cultivated in 3 organic farms. Agronomic traits are collected.



### 02 Pathogens Evaluation

Leaf and fruits samples are evaluated for the presence of EB and LB. Alternaria ifas are evaluated by microscope and by DNA sequencing to assess the true Alternaria strains diffused in northern Italy.



### 03 Processing Tests

Tomato fruits are processed into juice and evaluated for physical parameters to assess the quality of the product once processed.

Physical parameters evaluated on juice in 2021 trials (gap between the variety and the average)

Varietà	LB resistant	EB resistant	Agronomic value	EB field evaluation	pH	Acidity	Brix	Bx/ac	Bostw	Col L	A/B
Syrax	no	no	7	3	-0,05	0,55	0,21	-0,11	-0,73	0,09	-0,02
Aleatico	no	yes	6,5	2	0,06	0,47	0,36	-0,13	-0,51	-0,01	-0,03
Rulander	no	yes	7,5	2	0,04	0,51	0,01	-0,23	-1,23	0,16	-0,03
Cartizze	yes	no	8	4	-0,04		-0,20		-0,81	0,33	-0,11
Milbech	yes	no	7,5	3	-0,07	0,48	0,09	-0,33	0,19	-0,08	-0,15
TS1001	yes	no	5	5	0,08	0,41	0,23	1,32	2,77	-0,06	-0,08
Gavis	no	no	5,5	2	0,03	0,40	-0,51	-0,65	2,49	-0,37	-0,01
Mascalì	no	yes	8,5	1	0,01	0,42	-0,39	-0,14	-1,96	0,03	-0,13
17T5441	no	no	6,5	4	-0,08	0,53	-0,04	-0,49	1,42	0,21	-0,10
17T5560	no	yes	6	1	0,08	0,47	-0,23	-0,77	1,34	0,48	0,01
Frecciarossa	yes	no	5	3	0,10	0,41	-0,21	-0,15	-0,98	-0,79	0,31
17T5473	yes	no	4,5	2	0,09	0,42	0,07	0,81	0,91	0,16	-0,17
Castelaner	no	yes	7,5	1	0,08	0,43	0,16	0,56	-0,58	-0,57	0,08
16T5414	yes	no	6		0,00		-0,32		0,15	0,35	0,09
20T6316	yes	no	5,5		-0,01		-0,47		-3,25	0,08	0,09

### 04 Early Blight Artificial Infection

A method for artificial infection of tomato plantlets by EB is required so many different techniques are evaluated.

Variety	% of infected leaves	Variety	% of infected leaves
Syrax	40,00%	Frecciarossa	45,00%
Aleatico	47,00%	17T5473	
Rulander	46,00%	Castelaner	49,00%
Cartizze	21,00%	16T5414	
Milbech		20T6316	
TS1001		21T6576	36,00%
Gavis	7,00%	21T6506	54,00%
Mascalì	27,00%	21T6504	32,00%
17T5441		19T5940	60,00%
17T5560	58,00%	20T6406	

### 05 Early Blight Toxins Evaluation

The evaluation of Alternaria toxins in tomato juice and puree is getting more and more important for processor, so a method to get fast and reliable indication has been developed.

**Ergosterol:** The sample has to be saponified with ethanolic potassium hydroxyd solution before to be extracted with petroleum ether. The extract is evaporated to dryness then redissolved in methanol and analyzed by HPLC on reversed- phase column with a PDA detector.

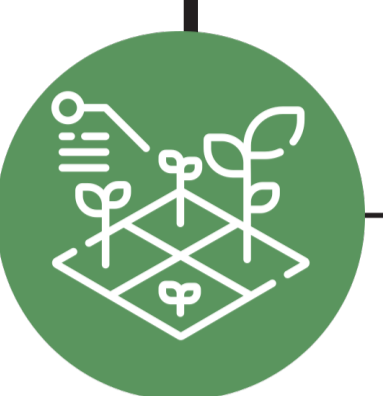
**Alternaria toxins:** The sample is extracted with acetonitrile acidified with 1% formic acid. MgSO4 and NaCl are added during the extraction phase and after freezing, the extract is analyzed by UHPLC-HRMS/MS.

### 06 Dissemination

An online course on organic tomato farming has been organized. A training trip at organic farms, tomato processor and CREA laboratories.



## 2nd YEAR



### 07 Trial Fields

The 4 best varieties are cultivated in large trials at 5 organic farms. Agronomic traits are collected.

Variety	Cycle	Vigour	Health.	Yield
- 20T6406 F1:	medium-late	mid-low	medium	medium
- CARTIZZE F1:	medium-early	medium	good	medium
- MASCALI F1:	medium	high	good	good
- MILBECH F1:	late	high	good	medium
- H5108 F1 (check):	early	low	medium	medium

Yields and healthiness data are still in progress

### 08 Pathogens Evaluation

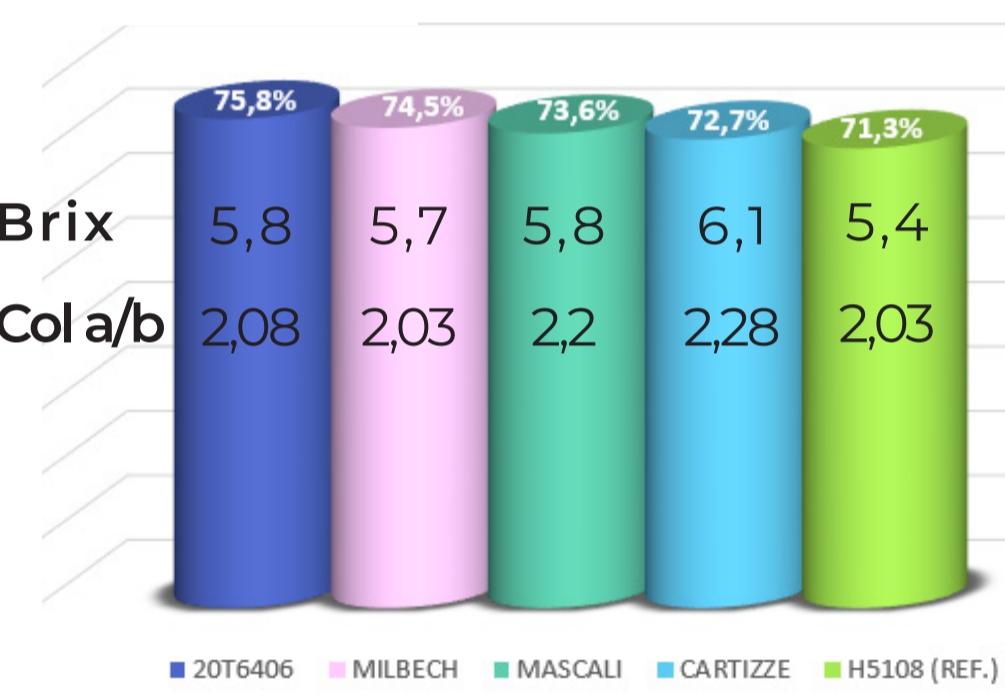
Leaf and fruits samples are evaluated for the presence of EB and LB. Alternaria ifas are evaluated by microscope and by DNA sequencing to assess the true Alternaria strains diffused in north of Italy.

### 09 Processing Tests

Tomato fruits are processed in plants that simulate the industrial process. Finally, juice, puree and dicing are obtained by each varieties. Physical and chemical parameters are evaluated to have indications on the quality and on the yield of the obtained products. The data will be repeated even after 6 months of storage.

VARIETY	FRESH FRUITS				JUICE										
	Fruit weight g	Pulp thickness mm	*Bx	Dry content (g/100g)	total sugars (g/100g)	RZ%	Acidity (g/100g)	RA% (Acidity/RS)	pH	Bw (cm/30sec)	color L	color a/b	Alternaria toxins (ug/Kg)	Ergosterol (mg/kg)	Lycopene (mg/100g)
20T6406	70	8,5	5,62	6,14	2,77	45,19%	0,51	8,19	4,21	16,5	25,5	2,36	n.d.	<0,2	8,7
CARTIZZE	60	5,8	6,01	6,46	2,87	44,41%	0,55	8,39	4,23	17,5	25,2	2,52	n.d.	<0,2	10,9
H5108 check	61	6,0	5,14	5,51	2,19	39,75%	0,53	9,51	4,24	20,0	25,5	2,26	n.d.	<0,2	8,7
MASCALI	77	9,0	6,31	6,72	3,33	49,37%	0,51	7,63	4,27	19,5	25,4	2,57	n.d.	<0,2	12,5
MILBECH	69	7,5	5,65	6,14	2,69	43,80%	0,59	9,61	4,16	17,5	26,6	2,31	n.d.	<0,2	9,6

### Dice



### Puree

VARIETY	*Bx	pH	Acidity (g/100g)	RA (Acidity/Bx)	color L	color a/b	Taste
20T6406	10,2	4,2	0,82	8,0	25,43	2,59	Dense and nice coloured, acid taste
CARTIZZE	9,9	4,16	0,76	7,7	25,01	2,71	Nice color and texture, very good taste
H5108 TEST	9,7	4,27	0,96	9,9	25,43	2,48	Nice color and texture but very acid taste
MASCALI	9,8	4,34	0,66	6,7	24,87	2,77	Bright colour, liquid texture but very good taste as sweet
MILBECH	9,7	4,14	0,97	10,0	25,72	2,43	Good texture, pale color, acid taste



### 10 Contaminants Evaluation

The juices are tested even for the presence of chemicals contaminant, toxins from fungus or bacteria and specific Alternaria toxins.

### 11 EB Resistance Tests

Plantlets and fruits are tested for resistance to artificial infection by EB.



### 12 Next steps

LCA (Life Cycle Assessment) analysis by examining the entire crop production process ( fertilization, water consumption, weed control, defense). Analysis of organic tomato production costs at field stages. **Publications** on specialized journals and the organization of a **final meeting** will help in spreading the results obtained during the 2 years project.

**DISCUSSION:** The Va.Po.Re. project has the aim to support organic farming by developing new healthier and better performing varieties both in the field and at the processor compared to the varieties developed for traditional farming. Other goals are: monitoring the fungal races present in Italy, developing techniques to evaluate the resistance to EB and to detect the presence of Alternaria toxins into tomato juice, quantifying the environmental and economic impact and of organic cultivation of processing tomato (to be done). Finally, we want the farmers and the processors know the results of these activities by organizing meetings in field and in-house, publishing article on specific journals as well as organizing courses and training trips.

At the moment the project is ongoing and will end in March 2023. Some activities are not completed yet, while some others are done. The 2021 monitoring of the Alternaria races has showed that the main race present in Northern Italy is similar to Alternaria solani if observed by optic microscope, while its genetic sequence appears closer to Alternaria alternata. The 2022 data will help clarifying the situation.

A new method has been developed for the artificial EB infection of tomato plantlets. A rapid and cheap technique based on UHPLC-HRMS/MS for the detection and quantification of Alternaria toxins has been established. During the second year trial the fields were poorly affected by EB so all the tomato juices are free from Alternaria toxins. The ergosterol analysis (to be done) will give indication on the healthiest tomato varieties.

The first year trial led to select 4 varieties for the following year but other varieties performed well and we hope to organize larger trial in the future even for these varieties: Rulander, Aleatico, Syrax and Castelaner.

The second year trial gave detailed information on the 4 varieties even if the final results will be available six months after the tomato products are stored. The selected varieties resist to EB (Mascalì) or to LB (Cartizze and Milbech) or both (20T6406). Even if these results come from one year trial at laboratory scale, at the moment we have obtained useful indications on the right variety to choose for organic farming depending on the final tomato product (juice, puree, paste or dice).

**20T6406 F1:** Suitable for juice, puree and dice  
**CARTIZZE F1:** Great quality and yield for puree and paste products

**MASCALI F1:** Excellent dice and paste. High lycopene content  
**MILBECH F1:** Excellent dice and good puree

**Acknowledgment:** Delta bio, Azienda agricola Buttini, ForB. This study was funded by the Emilia-Romagna Region within the Rural Development Plan 2014–2020 Op. 16.1.01– GO EIP-Agri- FA 3A