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# CHITOSAN, ESSENTIAL OILS AND OZONE AS TOOLS FOR THE MANAGEMENT OF POSTHARVEST DECAY OF FRESH FRUIT

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 *GianfRomanazzi*

*Ancona, 23 January 2024*



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d'Investigacions Agràries



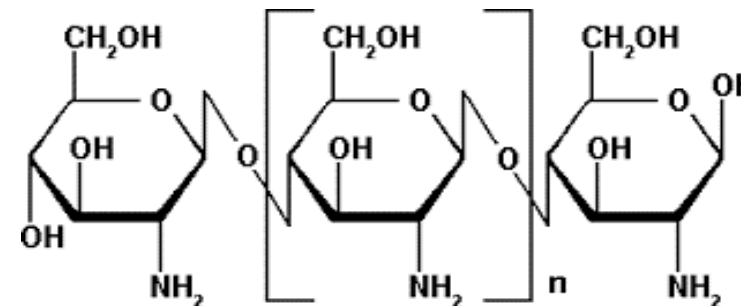
**DECCO**  
Naturally Postharvest

## Chitin related food science today (and two centuries ago)



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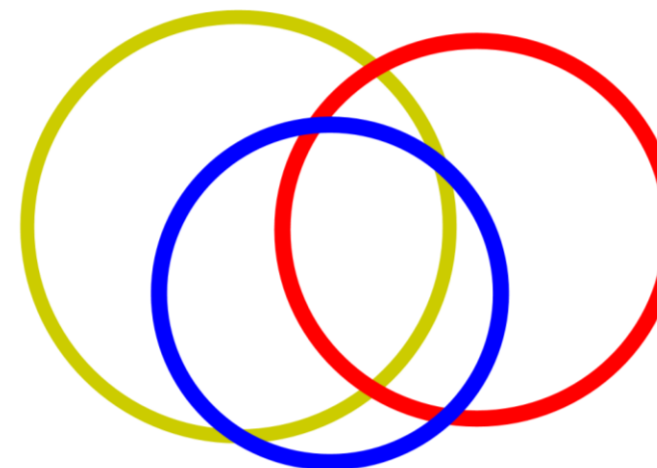


## Chitosan, a Biopolymer With Triple Action on Postharvest Decay of Fruit and Vegetables: Eliciting, Antimicrobial and Film-Forming Properties

Gianfranco Romanazzi<sup>1\*</sup>, Erica Feliziani<sup>1</sup> and Dharini Sivakumar<sup>2</sup>

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**Antimicrobial  
properties  
(35-45%)**



**Eliciting  
properties  
(30-40%)**

**Film-forming properties  
(20-30%)**



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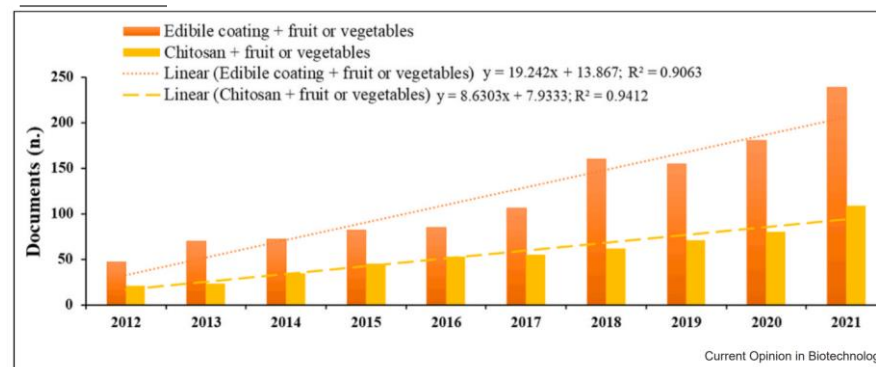
## Chitosan and other edible coatings to extend shelf life, manage postharvest decay, and reduce loss and waste of fresh fruits and vegetables

Gianfranco Romanazzi and Marwa Mourni

Current Opinion in Biotechnology 2022, 78:102834

<https://doi.org/10.1016/j.copbio.2022.102834>

Figure 2



Number of documents available on Scopus through searches with keywords "edible coating and fruit or vegetables; chitosan, postharvest or post-harvest and fruit or vegetables" in "Article title, Abstract, and Keywords" published over the last 10 years (Source: Scopus, accessed on 26 June 2022; <https://www.scopus.com>).

Table 2

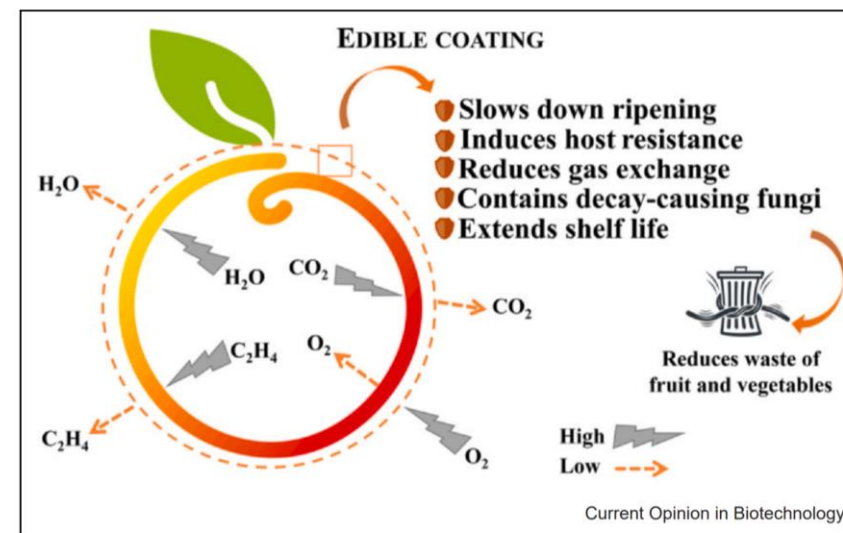
Examples of chitosan-based commercial products that are available for the control of diseases of fresh fruits and vegetables (modified by Romanazzi et al. [17]).

Product trade name	Company (country)	Formulation	Active ingredient (%)
Chito plant	ChiPro GmbH (Bremen, Germany)	Powder	99.9
Chitosano	Agrilaete (Palmanova, UD, Italia)	Powder	100
Chitosano denso		Liquid	50
OII-YS <sup>a</sup>	Venture Innovations (Lafayette, LA, USA)	Liquid	2
KaitoSol	Advanced Green Nanotechnologies Sdn Bhd (Cambridge, United Kingdom)	Liquid	12.5
Armour-Zen	Botry-Zen Limited (Dunedin, New Zealand)	Liquid	14.4
Biorend	Bioagro S.A. (Chile)	Liquid	1.25
Kiforce	Alba Milagro (Milano, Italy)	Liquid	6
FreshSeal	BASF Corporation (Mount Olive, NJ, USA)	Liquid	2.5
ChitoClear	Primex ehf (Siglufjordur, Iceland)	Powder	100
Bioshield	Seafresh (Bangkok, Thailand)	Powder	100
Biochikol 020 PC	Gumitex (Lowics, Poland)	Liquid	2
Kadozan	Lytone Enterprise, Inc. (Shanghai Branch, China)	Liquid	2
Kendal Cops	Valagro (Atessa, CH, Italy)	Liquid	4
Mastgrape	Enoceca (Vegrar, VR, Italy)	Liquid	5
Prevatect	Ascenza (Saronno, VA, Italy)	Liquid	5
Chitosano Serbios	Serbios (Badia Polesine, RO, Italy)	Liquid	5
Chitosano	Bioplanet Srl (Cesena, Italy)	Liquid	1.9
Chitosano DC	Dal Cin Gildo Spa (Concorezzo, MB, Italy)	Liquid	2
Ibisco <sup>b</sup>	Gowan Italia s.r.l. (Faenza, RA, Italy)	Liquid	15

<sup>a</sup> Contains 6% yucca extract.

<sup>b</sup> The formulation is based on an average of 12.5% of COS (chito-oligosaccharides)-OGA (oligo-galacturonides), with a chitosan concentration of 15%.

Figure 1



Main proprieties of edible coatings applied on fruits and vegetables, affecting the permeability to ethylene ( $C_2H_4$ ), water ( $H_2O$ ), oxygen ( $O_2$ ), and carbon dioxide ( $CO_2$ ).

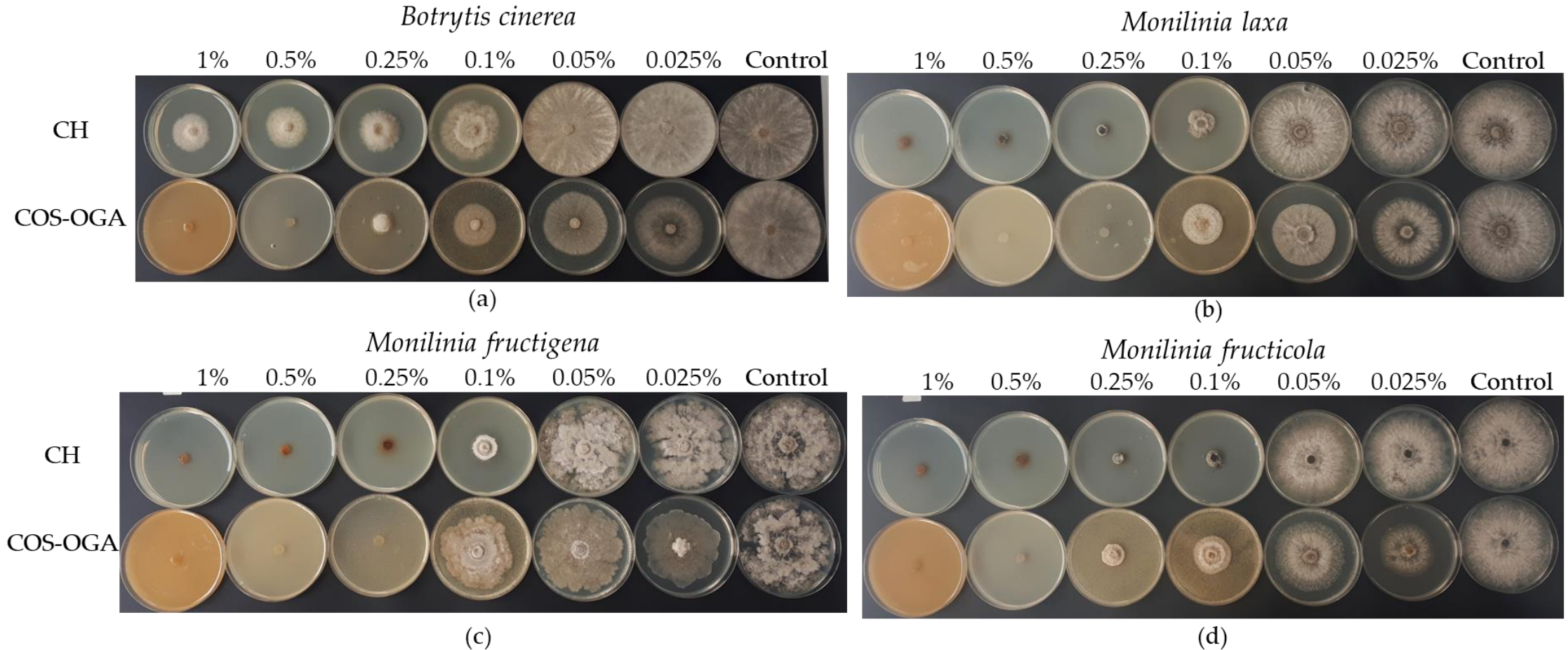
## Effectiveness of chitosan and COS-OGA against postharvest pathogens

- *In vitro* assays were conducted to evaluate mycelial growth inhibition of CH and COS-OGA against *Monilinia laxa*, *M. fructigena*, *M. fructicola*, and *Botrytis cinerea*
- CH (100%) and COS-OGA (1.25%) treatments at **1%**, **0.5%**, **0.25%**, **0.1%**, **0.05%** and **0.025%** concentrations were prepared in PDA and inoculated with 7-day old mycelia plugs

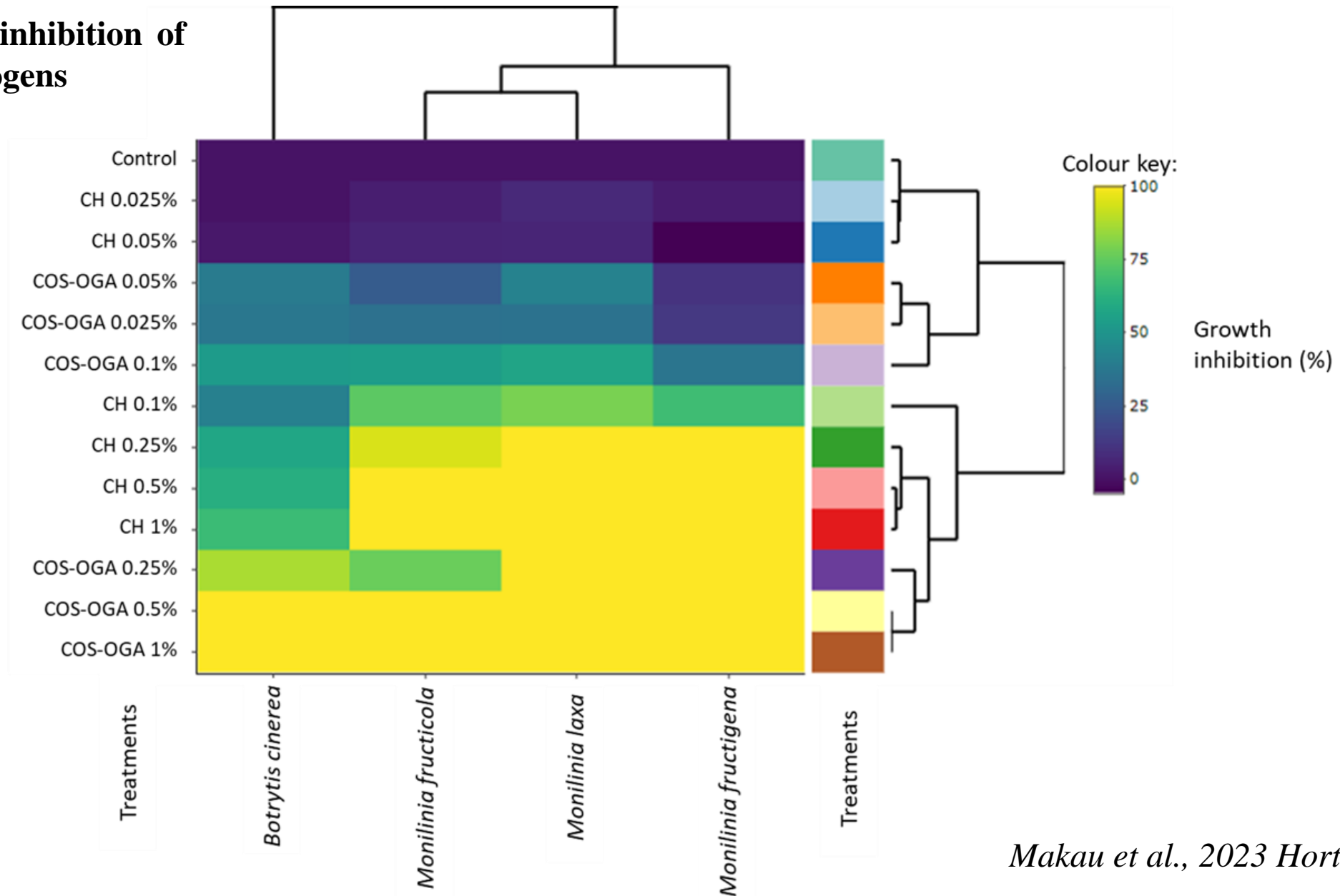


- Petri dishes were incubated at  $20 \pm 1^\circ\text{C}$  and mycelia growth was monitored daily
- Inhibition data were subjected to analysis of variance (ANOVA) using SPSS software

# Mycelial growth of postharvest pathogens



## Mycelial growth inhibition of postharvest pathogens



Communication

## Effects of Commercial Natural Compounds on Postharvest Decay of Strawberry Fruit

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\* Correspondence: g.romanazzi@univpm.it

<sup>†</sup> These authors contributed equally to this work.

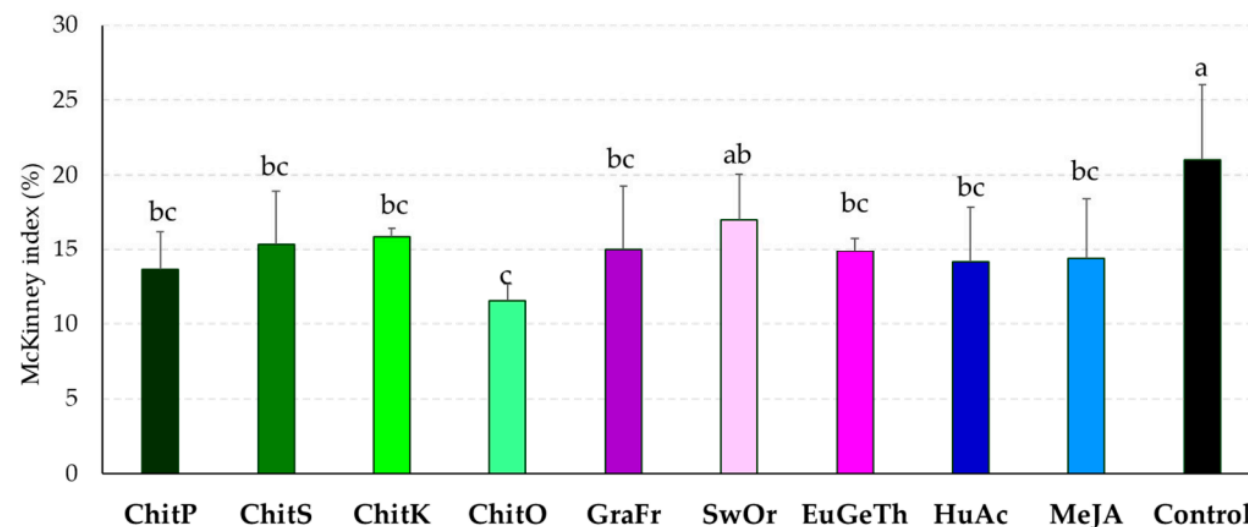
**Abstract:** Gray mold and Rhizopus rot, which is caused by *Botrytis cinerea* and *Rhizopus stolonifer*, respectively, are the most destructive forms of postharvest decay of the strawberry fruit. In this work, we tested the effectiveness of the control on the postharvest decay of the strawberry fruit (*Fragaria × ananassa* Duch cv. 'Monterey') following postharvest applications of six commercial natural compounds: chitosan-based coating compound (1% of 'ChitP', 'ChitS', 'ChitK', 'ChitO'), commercial essential oil (EOs) products based on grapefruit seed extract (0.5% of 'GraFr'), sweet orange (0.5% of 'SwOr'), a product that included eugenol, geraniol, and thymol EO, (0.4% of 'EuGeTh'), an organic compound as humic acid (0.5% w/v of 'HuAc'), and, lastly, methyl jasmonate plant growth regulator (1% v/v 'MeJA'). Strawberries were dipped in solution for 30 s and incubated at room temperature (20 ± 0.5 °C) or at cold storage conditions (4 ± 0.5 °C) following 4 days of shelf life at 20 °C. The treatments with 'ChitP', 'ChitS', and 'ChitO' provided ~30%–40% reduction of gray mold in cold storage conditions, while the 'MeJA', 'SwOr', and 'GraFr' with high activities of volatile substances were more effective at controlling gray mold at room temperature. 'HuAc', 'ChitK', and 'ChitO' were more effective at controlling Rhizopus rot in both cold storage (~50%) and room temperature conditions.

**Keywords:** basic substances; *Botrytis cinerea*; *Rhizopus stolonifer*; strawberry



Citation: Rajestary, R.; Landi, L.; Romanazzi, G. Effects of Commercial Natural Compounds on Postharvest Decay of Strawberry Fruit. *Coatings* 2023, 13, 1515.

<https://doi.org/10.3390/coatings13091515>



**Figure 1.** McKinney's index of gray mold of the 'Monterey' strawberry fruit. Strawberries were treated after harvest, stored for 7 days at 4 ± 0.5 °C, and then exposed to 4 days of shelf life at 20 ± 1 °C and 95% to 98% relative humidity. Values with different small letters are different at  $p < 0.05$ . Note: 'ChitP' = Chito Plant powder; 'ChitS' = Chito Plant solution; 'ChitK' = KaitoSol; 'ChitO' = OII-YS; 'GraFr' = DF-100 Forte; 'SwOr' = Prev-Am plus; 'EuGeTh' = 3Logy; 'HuAc' = Humic acid; 'MeJA' = methyl jasmonate.

Communication

## Effects of Commercial Natural Compounds on Postharvest Decay of Strawberry Fruit

Razieh Rajestary <sup>†</sup>, Lucia Landi <sup>†</sup> and Gianfranco Romanazzi <sup>\*†</sup>

Department of Agricultural, Food and Environmental Sciences, Marche Polytechnic University, Via Brecce Bianche, I-60131 Ancona, Italy; sabasol2002@yahoo.com (R.R.); l.landi@univpm.it (L.L.)

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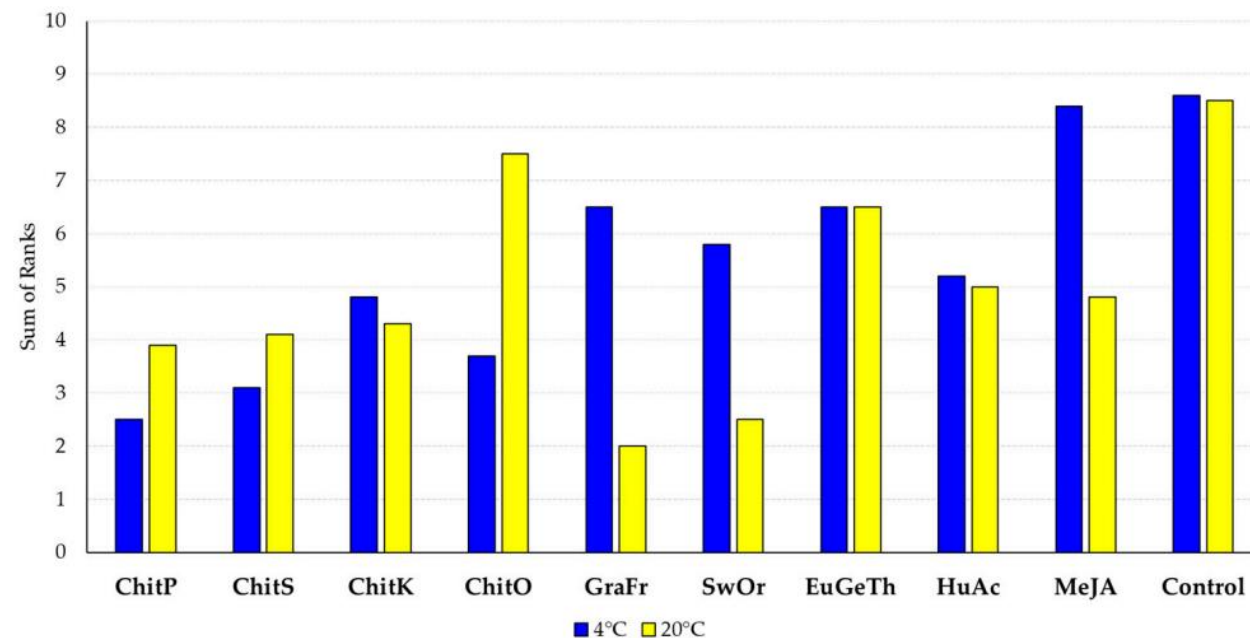
<sup>†</sup> These authors contributed equally to this work.

**Abstract:** Gray mold and Rhizopus rot, which is caused by *Botrytis cinerea* and *Rhizopus stolonifer*, respectively, are the most destructive forms of postharvest decay of the strawberry fruit. In this work, we tested the effectiveness of the control on the postharvest decay of the strawberry fruit (*Fragaria × ananassa* Duch cv. ‘Monterey’) following postharvest applications of six commercial natural compounds: chitosan-based coating compound (1% of ‘ChitP’, ‘ChitS’, ‘ChitK’, ‘ChitO’), commercial essential oil (EOs) products based on grapefruit seed extract (0.5% of ‘GraFr’), sweet orange (0.5% of ‘SwOr’), a product that included eugenol, geraniol, and thymol EO, (0.4% of ‘EuGeTh’), an organic compound as humic acid (0.5% w/v of ‘HuAc’), and, lastly, methyl jasmonate plant growth regulator (1% v/v ‘MeJA’). Strawberries were dipped in solution for 30 s and incubated at room temperature (20 ± 0.5 °C) or at cold storage conditions (4 ± 0.5 °C) following 4 days of shelf life at 20 °C. The treatments with ‘ChitP’, ‘ChitS’, and ‘ChitO’ provided ~30%–40% reduction of gray mold in cold storage conditions, while the ‘MeJA’, ‘SwOr’, and ‘GraFr’ with high activities of volatile substances were more effective at controlling gray mold at room temperature. ‘HuAc’, ‘ChitK’, and ‘ChitO’ were more effective at controlling Rhizopus rot in both cold storage (~50%) and room temperature conditions.

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Citation: Rajestary, R.; Landi, L.; Romanazzi, G. Effects of Commercial Natural Compounds on Postharvest Decay of Strawberry Fruit. *Coatings* 2023, 13, 1515.  
<https://doi.org/10.3390/coatings13091515>





**Figure 2.** The effect of postharvest treatment with natural compounds on the reduction of gray mold on strawberries according to rank analysis. The fruit was kept at 4 °C and 20 ± 1 °C, 95%–98% RH. Note: ‘ChitP’ = ‘Chito Plant powder; ‘ChitS’ = Chito Plant solution; ‘ChitK’ = KaitoSol; ‘ChitO’ = OII-YS; ‘GraFr’ = DF-100 Forte; ‘SwOr’ = Prev-Am plus; ‘EuGeTh’ = 3Logy; ‘HuAc’ = Humic acid; ‘MeJA’ = Methyl jasmonate.



Article

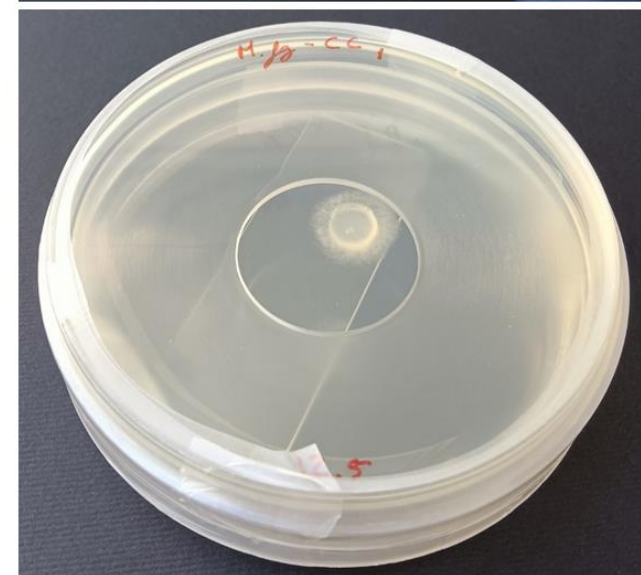
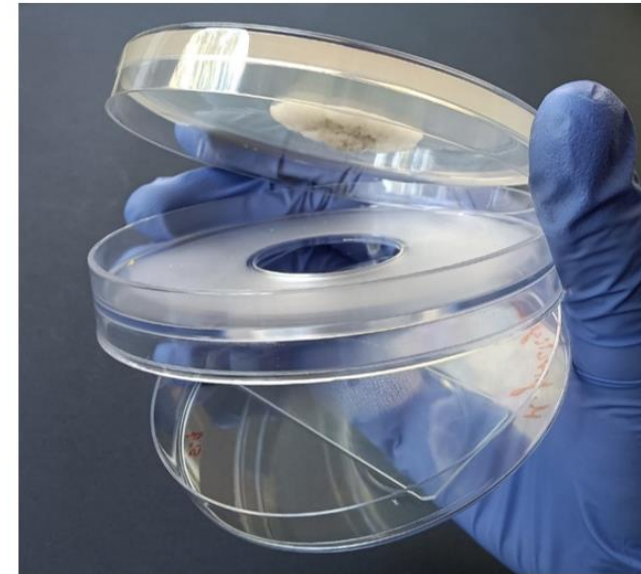
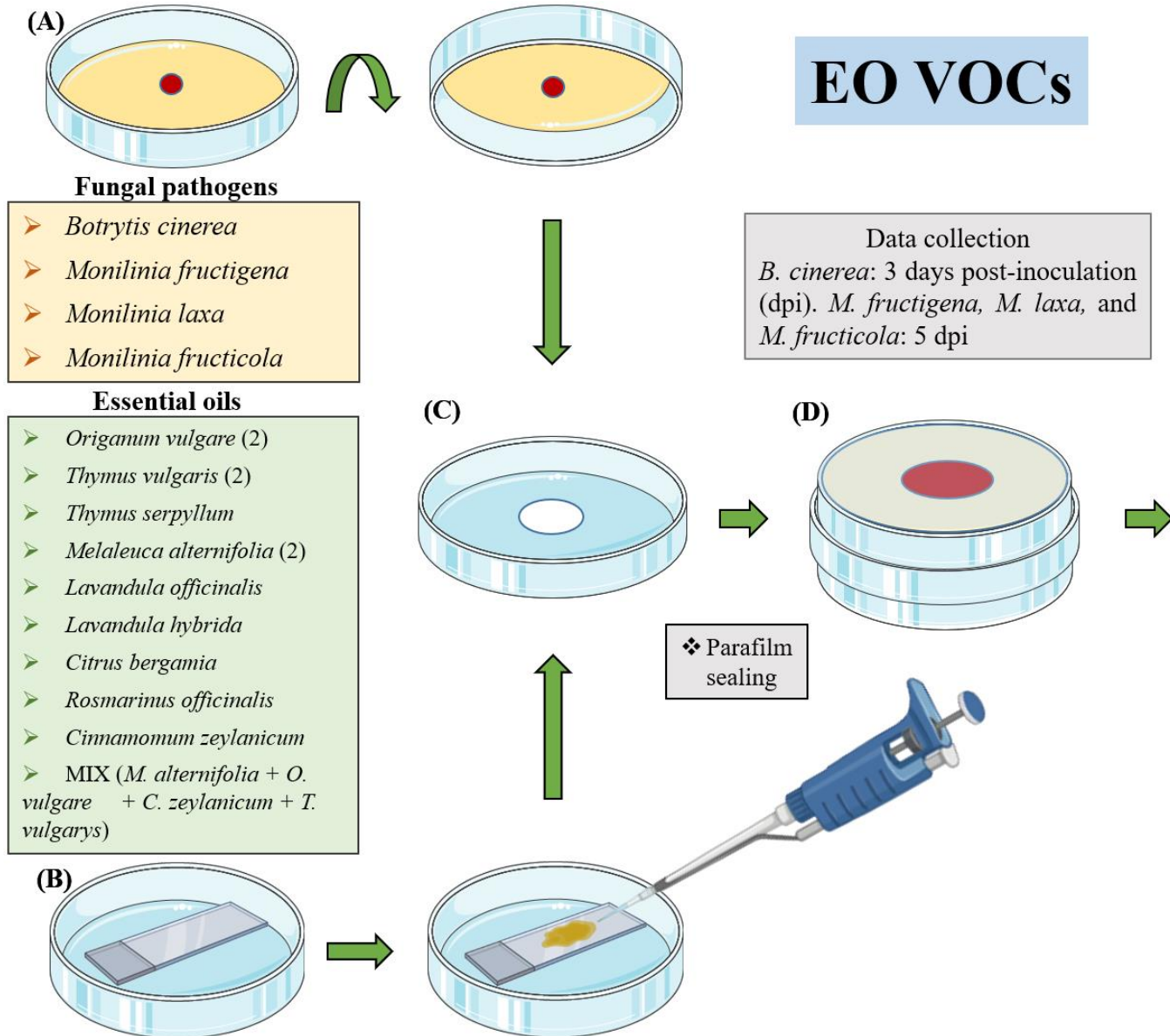
# Preharvest Application of Commercial Products Based on Chitosan, Phosphoric Acid Plus Micronutrients, and Orange Essential Oil on Postharvest Quality and Gray Mold Infections of Strawberry

Razieh Rajestary <sup>1,†</sup>, Panayiota Xylia <sup>2,†</sup>, Antonios Chrysargyris <sup>2</sup>, Gianfranco Romanazzi <sup>1</sup>  and Nikolaos Tzortzakos <sup>2,\*</sup> 

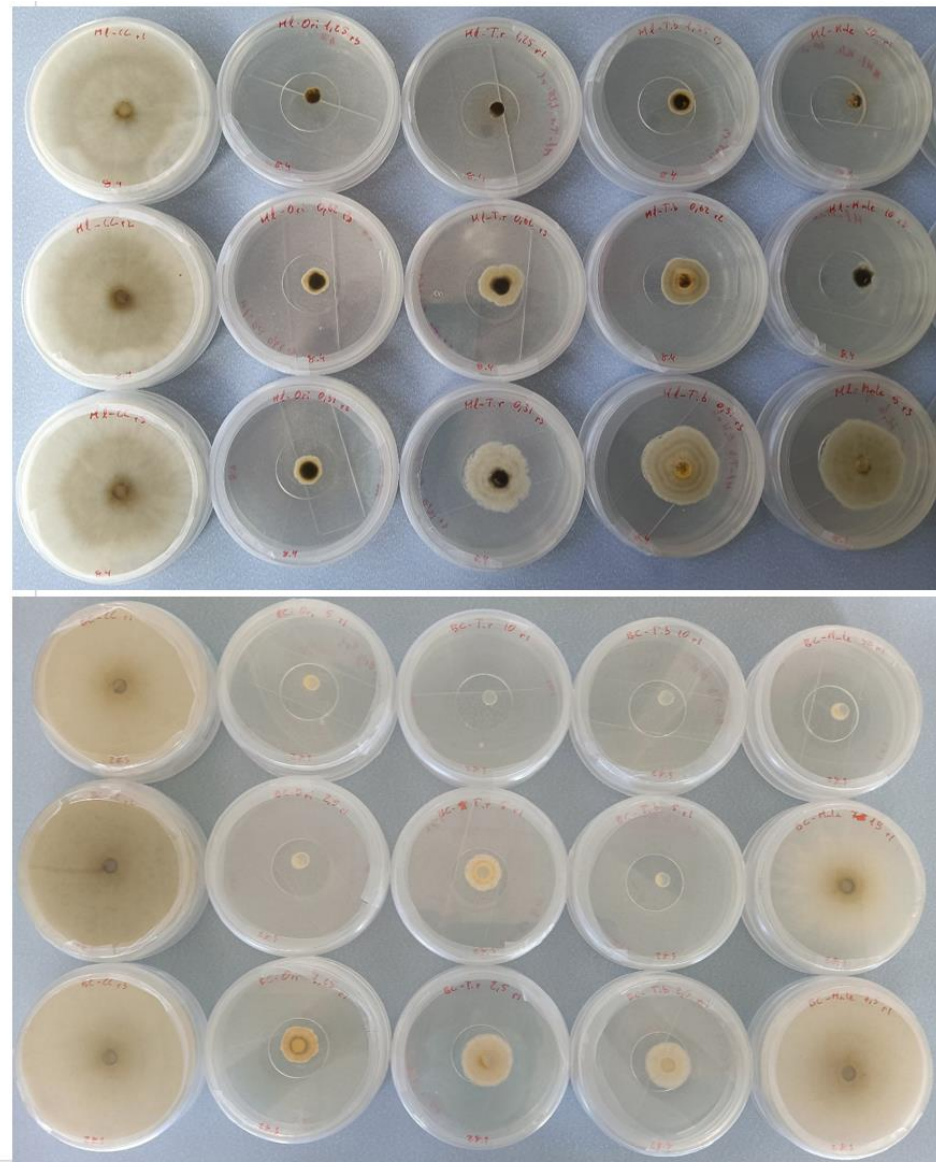
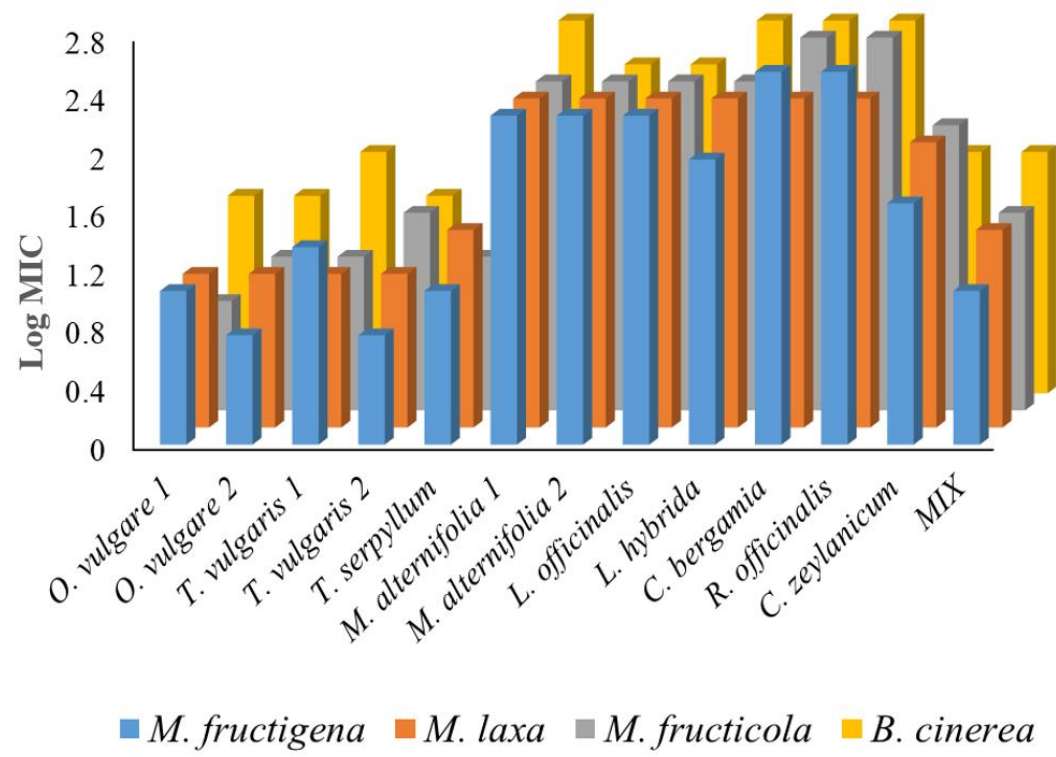
*No changes in fruit quality were recorded*

**Table 1.** McKinney index of gray mold recorded on strawberries treated with commercial natural compound formulations. The fruit were kept for 7 d at  $4 \pm 1$  °C, followed by 5 d shelf life at 20 °C and 95–98% RH. Different letters indicate that the values are significantly different according to a Tukey’s test ( $p \leq 0.05$ ).

Treatment	McKinney’s Index (%)
Control	55.71 ± 15.48 a
Chitosan	36.19 ± 9.11 b
Phosphoric acid plus micronutrients (PA+MN)	49.05 ± 13.57 ab
Sweet orange EO	49.52 ± 6.51 ab



**Minimum inhibitory concentration (MIC) of essential oil volatiles**



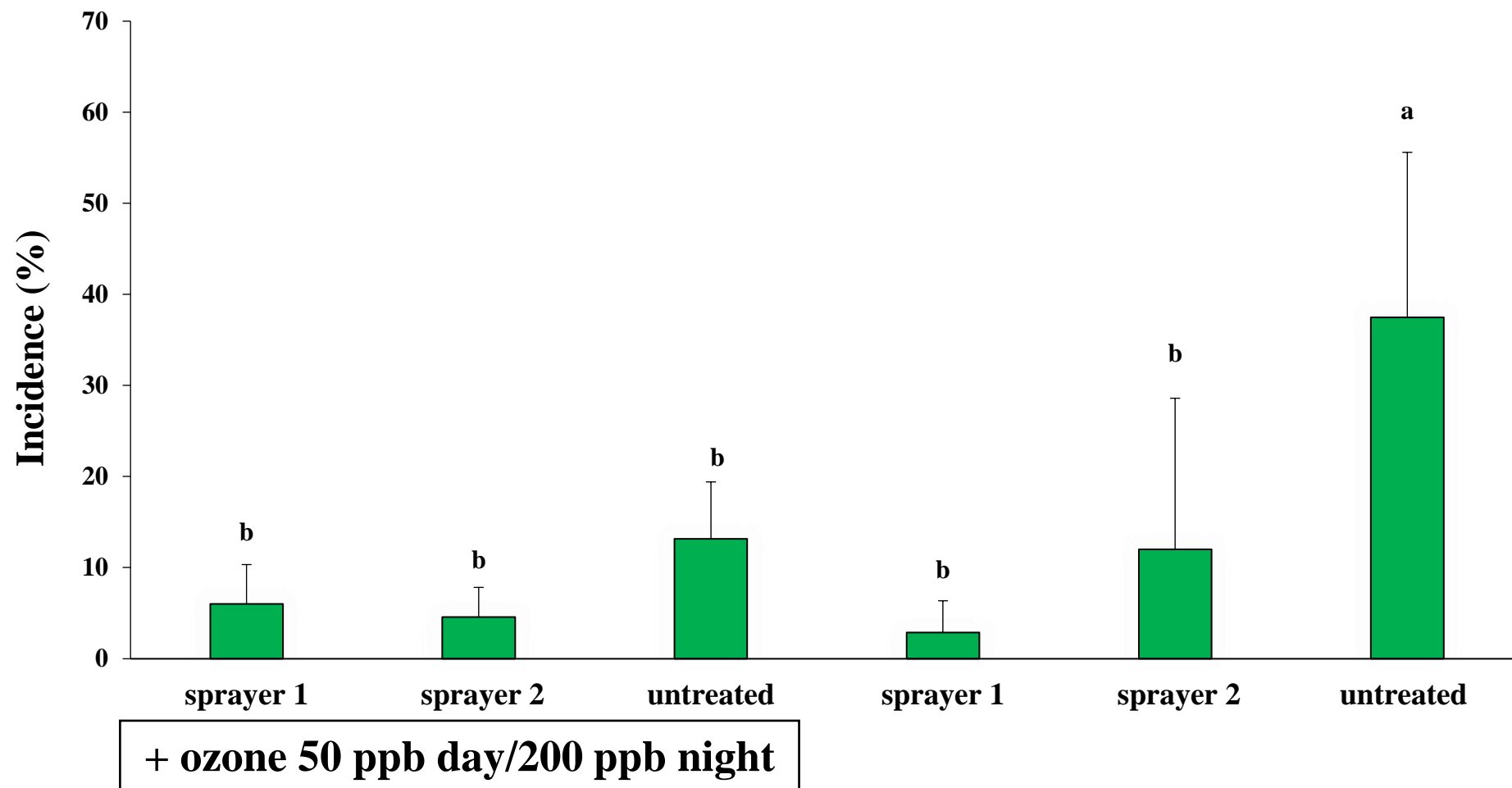
- **Preharvest treatment:** synthetic fungicides
- **Preharvest treatment:** gaseous ozone
- **Cultivar:** Extreme 486
- 10 boxes (35 fruits/box) per treatment
- **Ozone: 50 ppb day/200 ppb night x 20 d**
- **Storage: 4°C**

Preharvest treatments	Postharvest treatments
Sprayer 1	with ozone
Sprayer 2	
Untreated	
Sprayer 1	without ozone
Sprayer 2	
Untreated	



The shelf life was monitored for 20 days

## Brown rot on peaches stored 20 days at 4°C and exposed to 7 days shelf life



Sprayer 1:



Sprayer 2:



### Royal Summer

### Extreme 486

Ozone 45 ppb

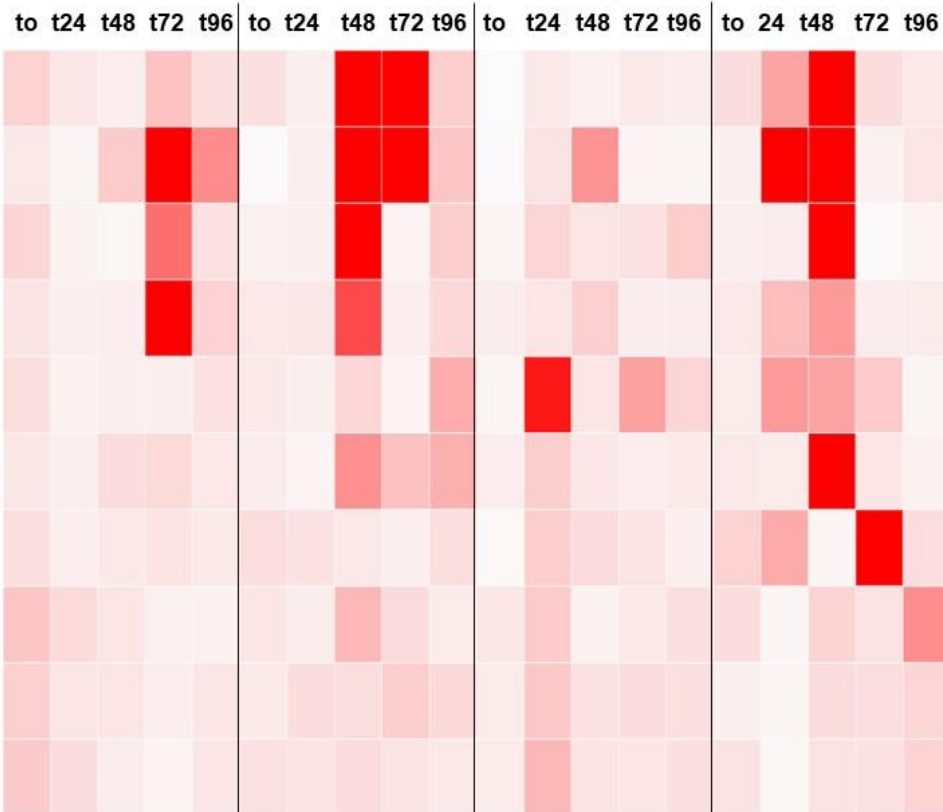
Ozone 50 ppb (day), 200 ppb (night)

10 days

20 days

10 days

20 days



1-aminocyclopropane-1-carboxylate oxidase (ACO)

major allergen (Pru av 1)

glutathione S-transferase (GST)

heat shock protein 90-5, chloroplastic (HSP90)

salicylic acid-binding protein 2 (SA)

ethylene responsive factor (ERF2)

cold shock protein (CSP)

jasmonic acid-amido synthetase (JAR1)

polygalacturonase (PG)

pathogenesis-related (PR) proteins-1

Mainly 20 day ozone treatment induced a rapid upregulation of ACO and ERF2 linked to ethylene production



Contents lists available at ScienceDirect

## Postharvest Biology and Technology

journal homepage: [www.elsevier.com/locate/postharvbio](http://www.elsevier.com/locate/postharvbio)Identification of volatile organic compounds as markers to detect *Monilinia fructicola* infection in fresh peachesBenedetta Fanesi<sup>a,1</sup>, Annamaria Lucrezia D'Ortenzio<sup>a,1</sup>, Anastasiya Kuhalskaya<sup>a</sup>,  
Ancuta Nartea<sup>a</sup>, Dennis Fiorini<sup>b</sup>, Marwa Moumni<sup>a</sup>, Lucia Landi<sup>a</sup>, Paolo Lucci<sup>a,\*</sup>,  
Gianfranco Romanazzi<sup>a</sup>, Deborah Pacetti<sup>a</sup><sup>a</sup> Department of Agricultural, Food and Environmental Sciences, Polytechnic University of Marche, Via Brecce Bianche, 60131, Ancona, Italy<sup>b</sup> School of Science and Technology, Chemistry Division, University of Camerino, Via Madonna delle Carceri 9/B, 62032 Camerino, Italy

## ARTICLE INFO

## Keywords:

Chromatography

Fungal pathogen

*Monilinia fructicola*

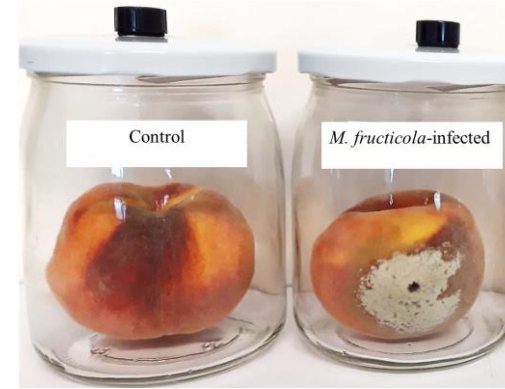
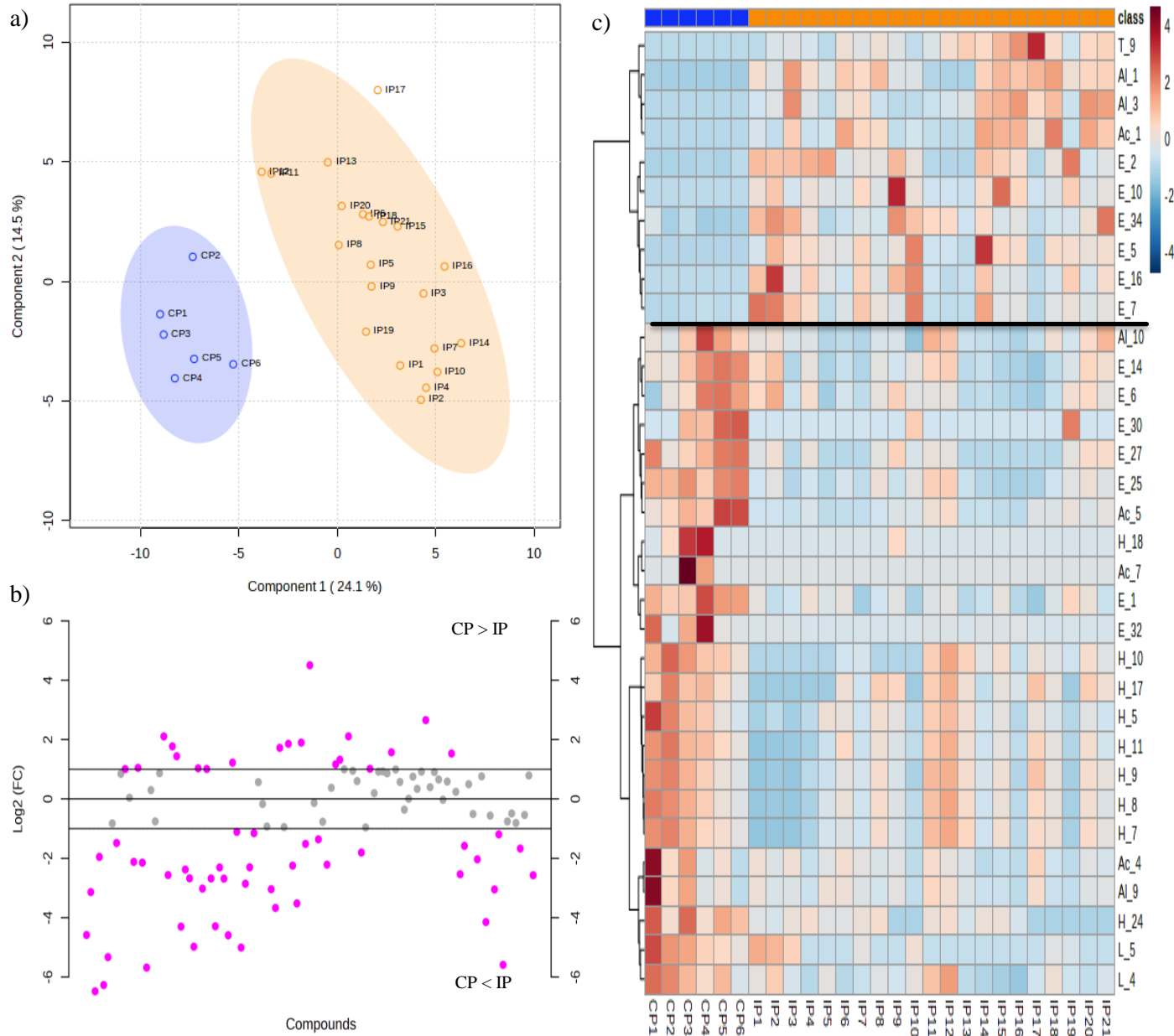
Peach

Sensors

Volatiles

## ABSTRACT

*Monilinia* spp. are among the main fungal pathogen affecting peaches, and they can cause severe pre- and postharvest losses. Development of smart packaging technologies (e.g., volatile indicators), facilitating infection detection and preventing other fruit from being contaminated, is still limited. In this study, we compared for the first time the aroma profile of whole healthy fresh peaches to *Monilinia fructicola*-artificially inoculated peaches, identifying discriminant volatile organic compounds (VOCs). More than one hundred VOCs were detected by applying head space solid-phase microextraction followed by GC-MS analysis. The level of methyl esters, hydrocarbons, lactones, and acids decreased in infected peaches indicating fruit aroma deterioration, while the concentration of ethyl esters and alcohols increased. In particular, the amount of ethanol and derived ethyl acetate reached a maximum of 24- and 20-fold increase in the infected peaches, respectively. Isobutanol, propyl acetate, and ethyl isovalerate were specifically emitted by *M. fructicola*-infected peaches. These compounds might serve as markers for the development of smart sensors allowing the detection of fungal infection.

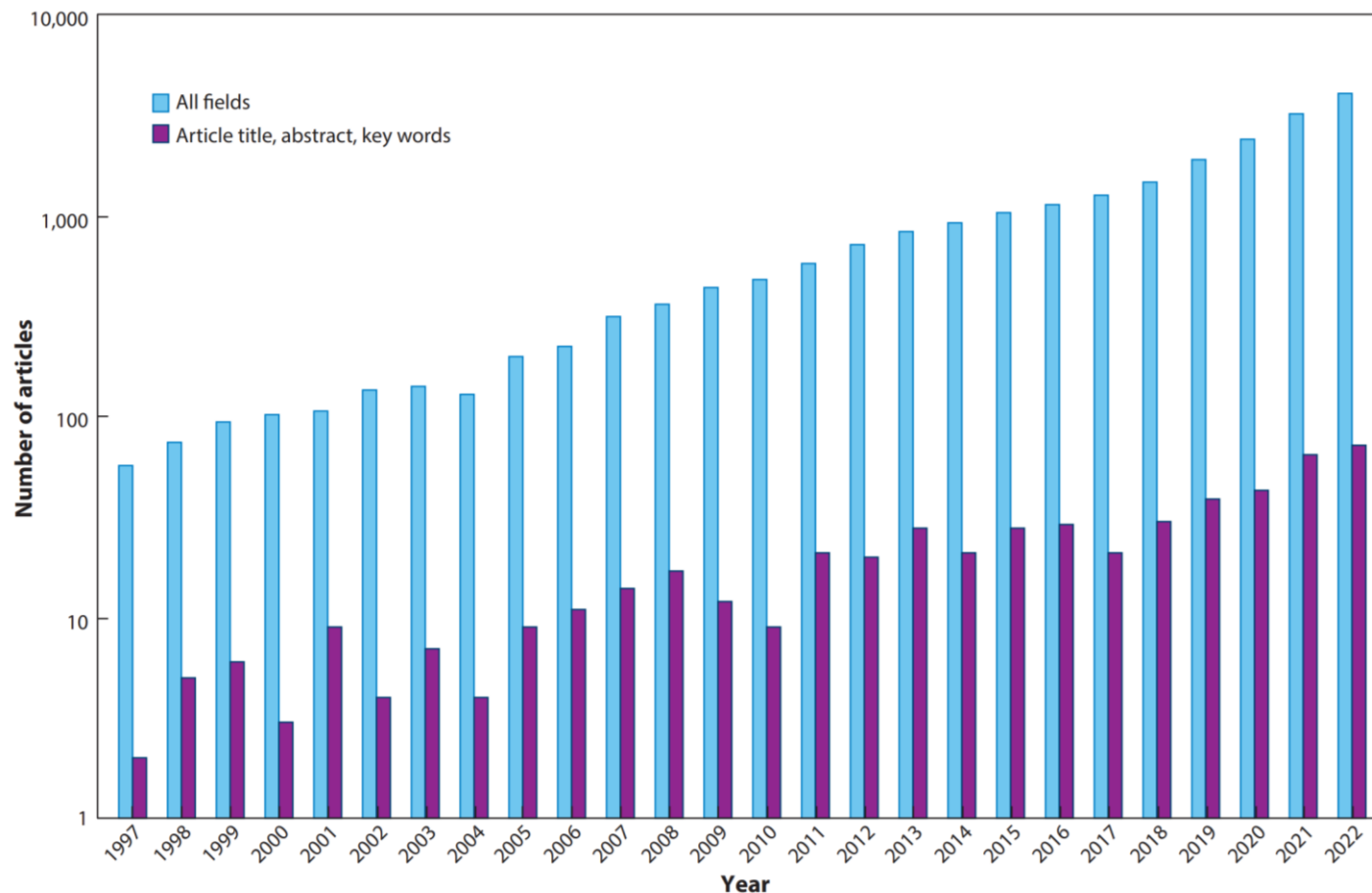


In cooperation with Deborah Pacetti, Food Technology Team, UNIVPM

VOCs variation between control (CP, blue) and *M. fructicola*-infected (IP, orange) peaches. (a) Partial least squares-discriminant analysis (PLS-DA) of VOC profile of CP (n=6) and IP (n=21). (b) Log<sub>2</sub> fold change with threshold 2, positive values indicating predominant VOCs in CP, negative values indicating VOCs accumulated in IP. (c) Hierarchical clustering heatmap of all CP and IP samples, showing the 33 statistically different ( $p < 0.05$ ) VOCs in the rows (letters and numbers are referred to Table 1). Cell colors indicate the relative percentage of the total volatile content for each VOC, blue representing low concentration and red high concentration. The black line separates the two clusters of compounds showing an opposite trend between CP and IP.



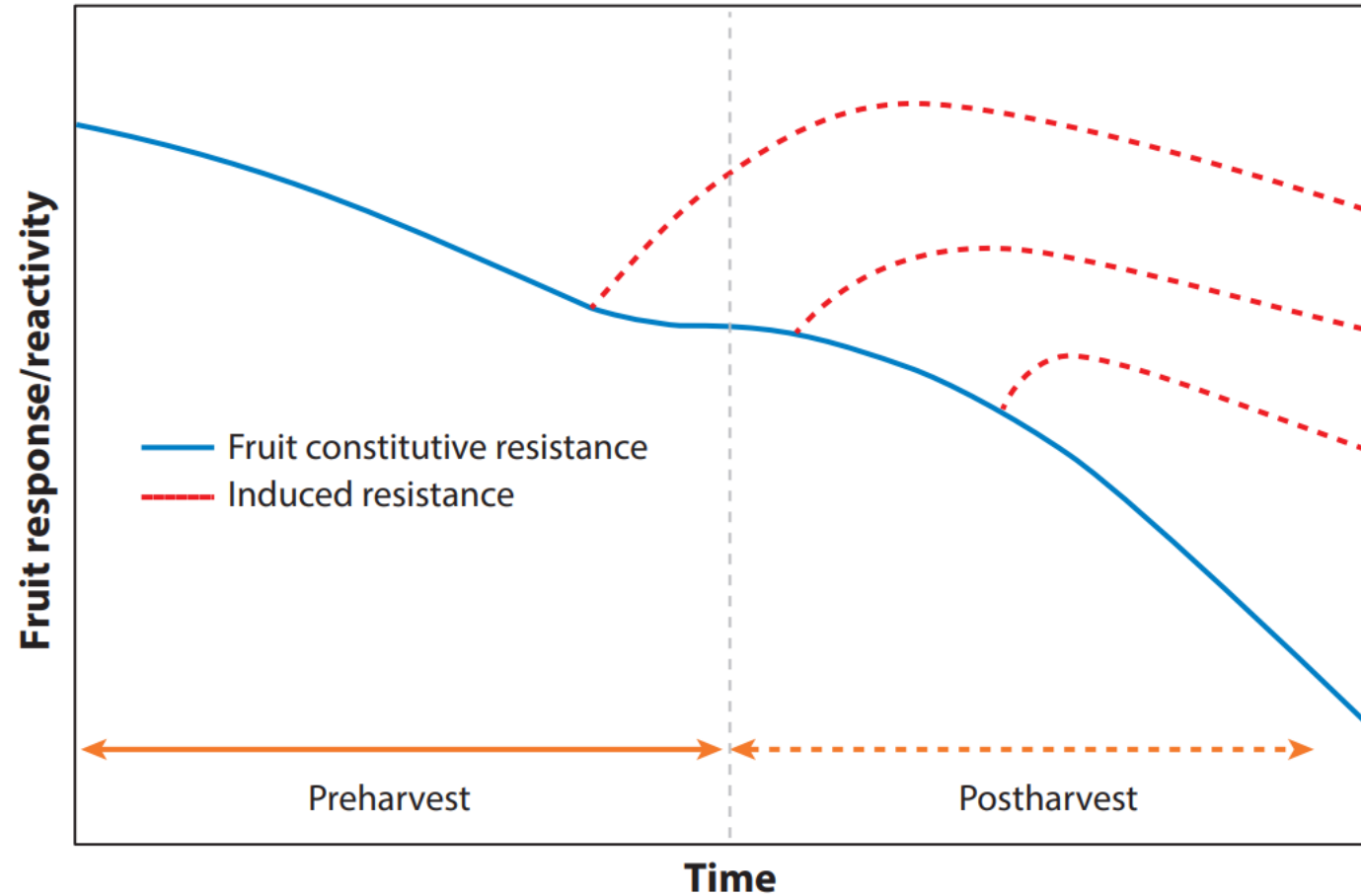
## Popularity of investigations on induced resistance



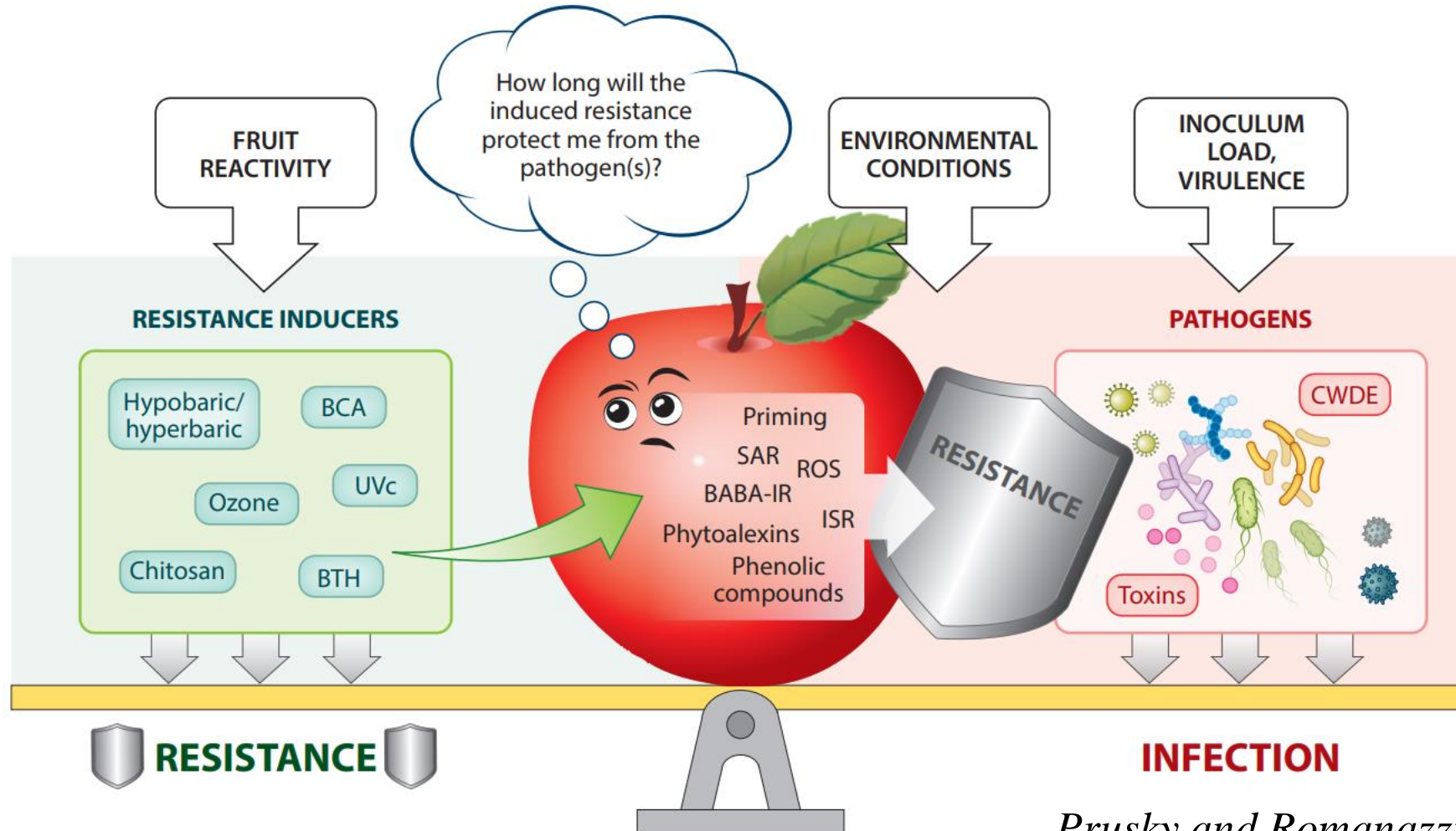
*Prusky and Romanazzi, 2023 ARP*

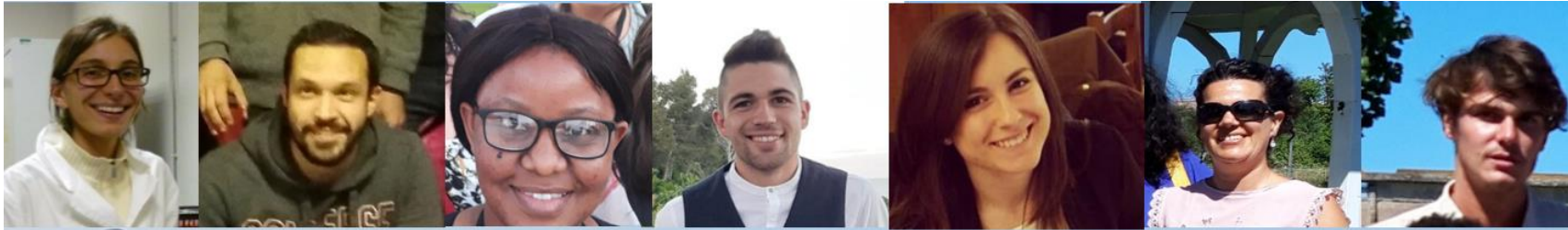
<https://doi.org/10.1146/annurev-phyto-021722-035135>

# Induced resistance in harvested fresh fruits



# Induced resistance in harvested fresh fruits





*Thanks  
for your  
attention*