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Meta-regression models describing the effects of essential oils and added lactic acid bacteria on *S. aureus* inactivation in cheese

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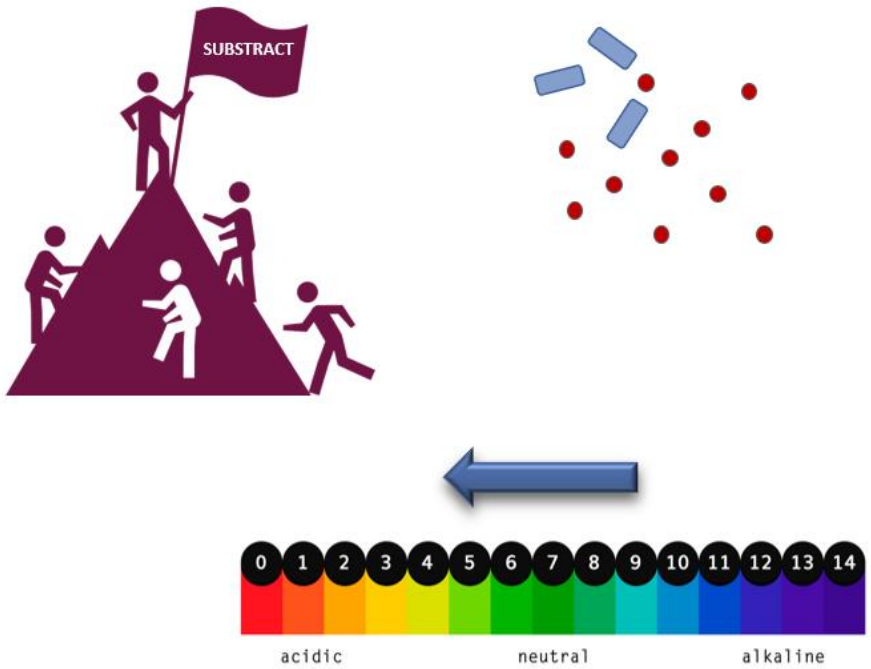
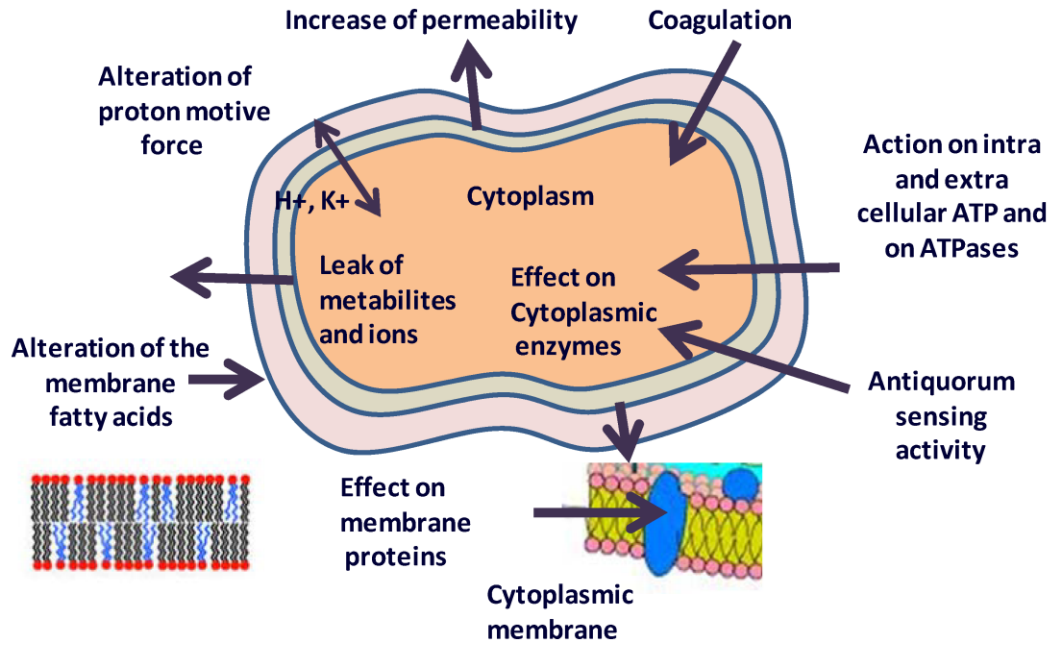
MOTIVATION

- **Biopreservatives** have been proposed as **hurdles** to increase microbiological safety of food products

Lactic acid bacteria (LAB)



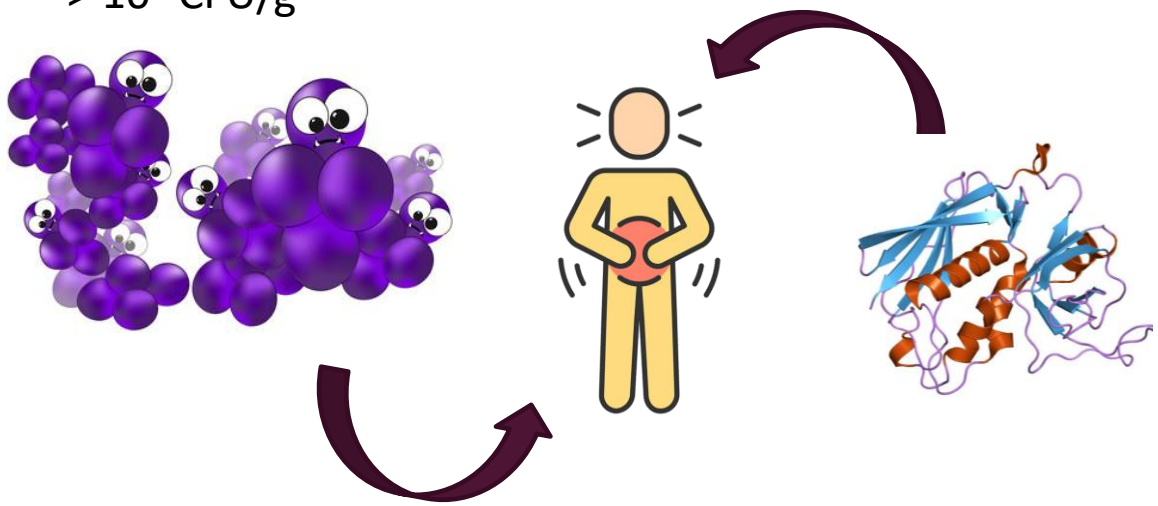
Essential oils (EO)



Adapted from Nazzaro et al., 2013

MOTIVATION

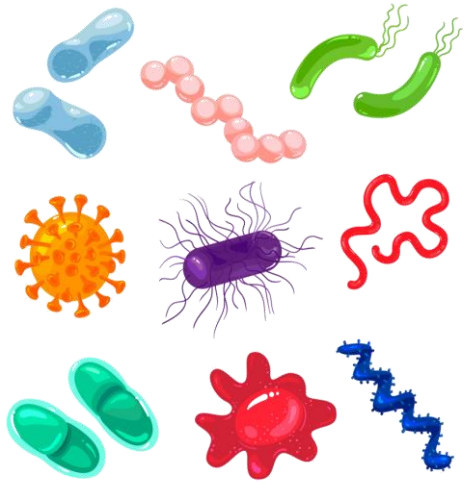
- Cheeses: non-satisfactory results in terms of pathogen contamination at retail level (EFSA and ECDC, 2018)
- *S. aureus* can be found in milk and dairy products, such as fermented milk and cheese



Important contamination issue!

MOTIVATION

- Available literature describing the effects of these biopreservation methods against several bacteria
- **Meta-analysis** and **meta-regression models** can be useful to understand pathogen growth, allowing optimisation of hurdles that provide long term stability and safety to cheeses



OBJECTIVES

To collect available literature on *S. aureus* inactivation in cheese containing added LAB and EOs

To harmonise the retrieved data by constructing two separate meta-regression models that summarise LAB and EOs effectiveness

METHODOLOGY

WEB OF SCIENCE
PubMed Scopus

Systematic literature search

Modelling in R Studio
(*nlme* package)

Mixed-effects linear models with weights

Population: cheeses with added EOs / LAB; Measured outcome: *S. aureus* mean log reduction

(i) Essential oils

Tested variables:
- Application type
- Storage temperature
- Inoculum level
- Application type × exposure time
- Application type × LnConcentration

$$\sqrt{R_{ik}} = (\beta_0 + u_i) + \beta_{1k} App_k + \beta_{2k}\{App_k \times t\} + (\beta_{3k} + v_i)\{App_k \times LnC\} + \beta_4 T + \beta_5 Inoc + \varepsilon_{ik}$$

(ii) Lactic acid bacteria

Tested variables:
- Application type
- Application type × exposure time

$$\sqrt{R_{ik}} = (\beta_0 + u_i) + \beta_{1k} App_k + \beta_{2k}\{App_k \times t\} + \varepsilon_{ikm}$$

Response variable:
√ Log Reduction
(log CFU/ml or /g)

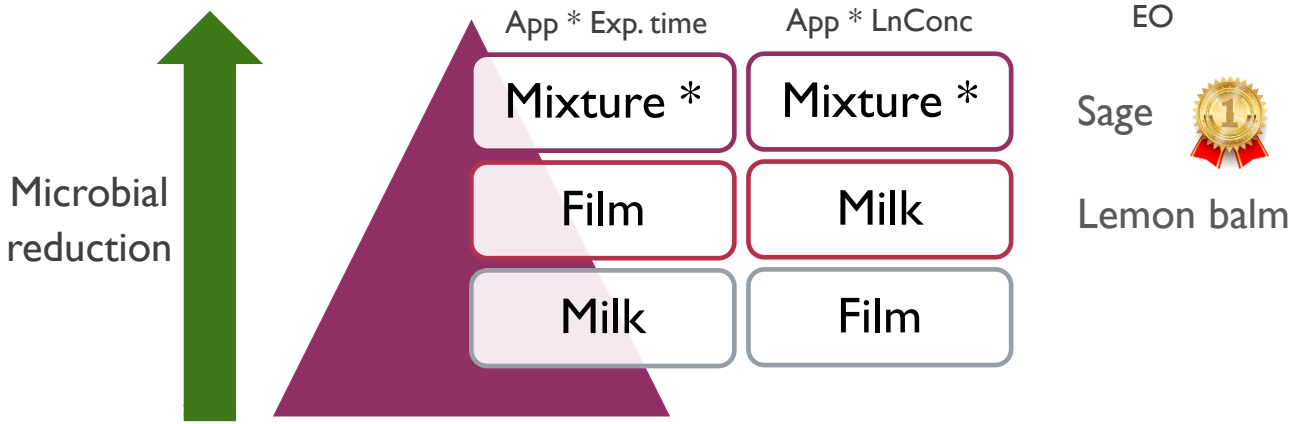
RESULTS

	Log reduction data		
	Levels	EOs (11 studies, N= 218)	LAB (11 studies, N= 81)
Application type	Mixture	58	11
	Film	83	0
	Milk	77	70
Exposure time (days)	[0, 30[	178	68
	[30, 60[	29	10
	[60, 75]	11	3
Storage temperature (°C)	[4, 15[	210	70
	[15, 35]	8	11
S. aureus inoculum level (log CFU/g or log CFU/ml)	[1.5, 3.5[	109	27
	[3.5, 5.5[	27	24
	[5.5, 8]	82	30
Antimicrobial concentration (log CFU/g, log CFU/ml, %v/v or %w/w)	[5×10^{-3}, 1.5[	168	38
	[1.5, 4.5[	50	0
	[4.5, 7.5[	0	4
	[7.5, 10]	0	39

RESULTS

(i) **EOs** meta-regression model

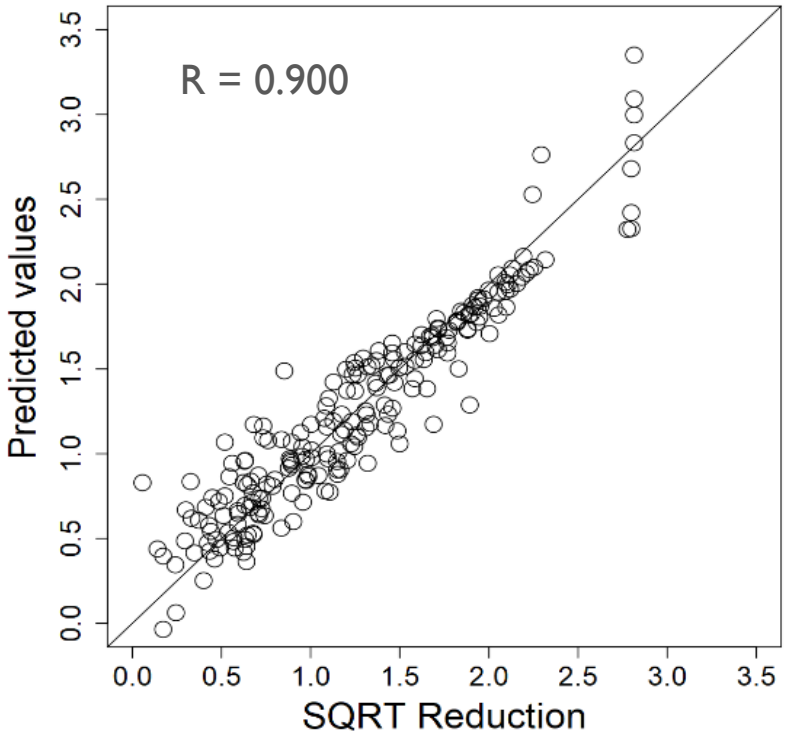
- Significant impact on pathogen inactivation:
  - ✓ Application type (p<.0001)
  - ✓ Application type * Exposure time (p<.0001)
  - ✓ Application type * Antimicrobial Conc. (p<.0001)
  - ✓ Inoculum level (p=0.019): negative association
- ✗ Storage temperature (p=0.246)



EO

Sage

Lemon balm



Heterogeneity analysis: > 95%

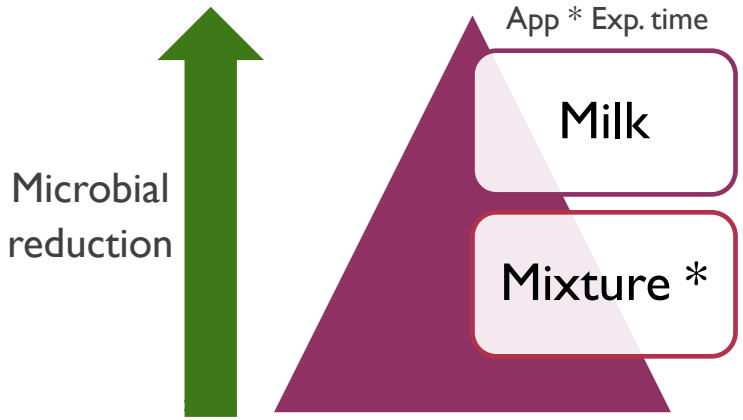
RESULTS

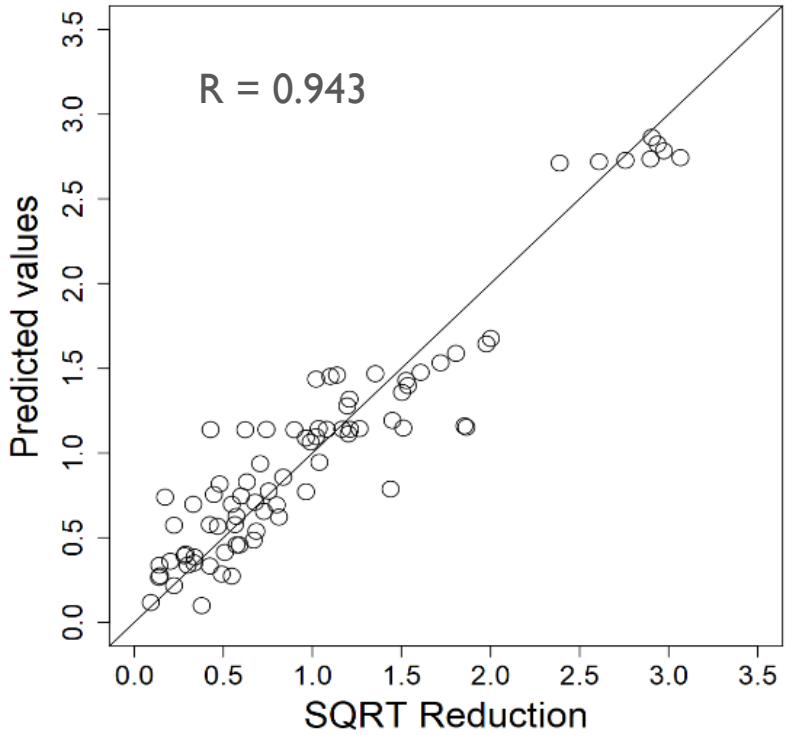
(i) **LAB** meta-regression model

- Significant impact on pathogen inactivation:

✗ Application type (p=0.091)

✓ Application type * Exposure time (p=0.040)





Heterogeneity analysis: ~12 %

CONCLUSIONS

- Antimicrobials' effectiveness depends on exposure time, application type, antimicrobial concentration…
- Insight on interactions between variables
- EO-embedded films lead to faster *S. aureus* inactivation
- Incorporation in cheese mixture is not an adequate practice
- Other sources of variability: type of milk, fermentation/ripening temperatures, single LAB strains vs. LAB-cocktails, …
- Effect of pathogen inoculum size: further research needed

CONCLUSIONS

Meta-regression modelling can be used for the

experimental design of challenge tests and

to optimise manufacturing processes and the use of biopreservatives!

→ **ensure microbial safety of cheeses** ←

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Cofinanciado por:

Thank you for your attention!

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