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## **Microbial spoilage of liquid egg products and its control by the detection of genetic or chemical markers**

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HAL Authorization



# Microbial spoilage of liquid egg products and its control by the detection of genetic or chemical markers



STLOpen Days  
19-21 March 2019



Sophie JAN  
MICROBIO Team



# Introduction



One of the leader in the production and consumption of eggs in the EU

- 14.9 billion eggs produced / year
- 213 eggs consumed /year / person

Itavi and AND International, 2017

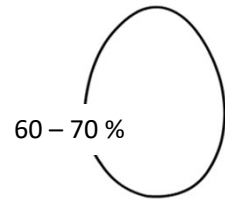
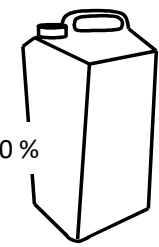
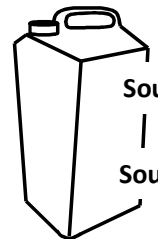


Table eggs



Egg products

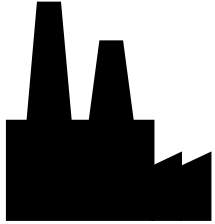
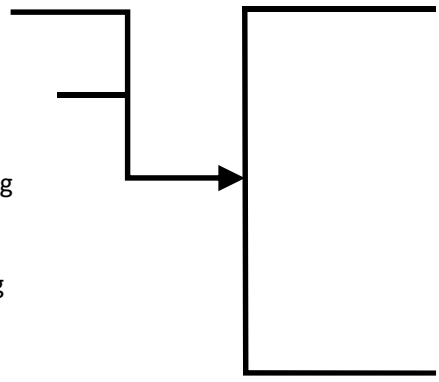


Source of nutrients

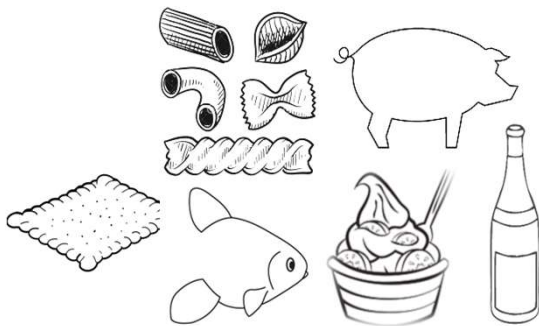
Source of ingredients

Egg products

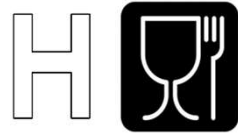
- Foaming
- Emulsifying
- Coloring
- Binding
- Thickening
- Flavoring



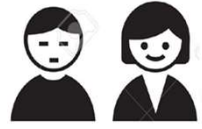
Food industry



Biscuits, pastries, desserts, pasta, chocolate and sugar confectionery, delicatessen, seafood products, wines, etc.



Hospitals  
Restaurants



Industrial catering and residential facilities



Food artisans

# What about egg microbiology?

Egg content

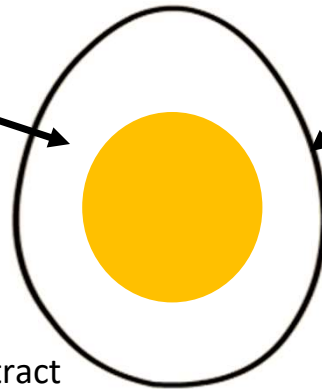
Sterile except under sporadic cases of contamination by *Salmonella* (mainly *Salmonella* Enteritidis) during egg formation

- Strong affinity for the avian reproductive tract
- Persistence on the eggshell surface
- Resistance to egg defence systems



Less than 1% of contamination but still 30% of strong evidence of salmonellosis caused by eggs or egg products in the EU

EFSA and ECDC, 2017



Eggshell surface

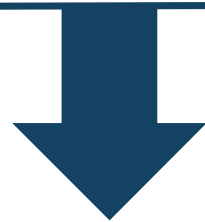
Contaminated in the environments of hen breeding and egg handling and storage

+++ Gram-positive bacteria

*Streptococcus, Aerococcus, Micrococcus, Staphylococcus, and low occurrence of Bacillus*

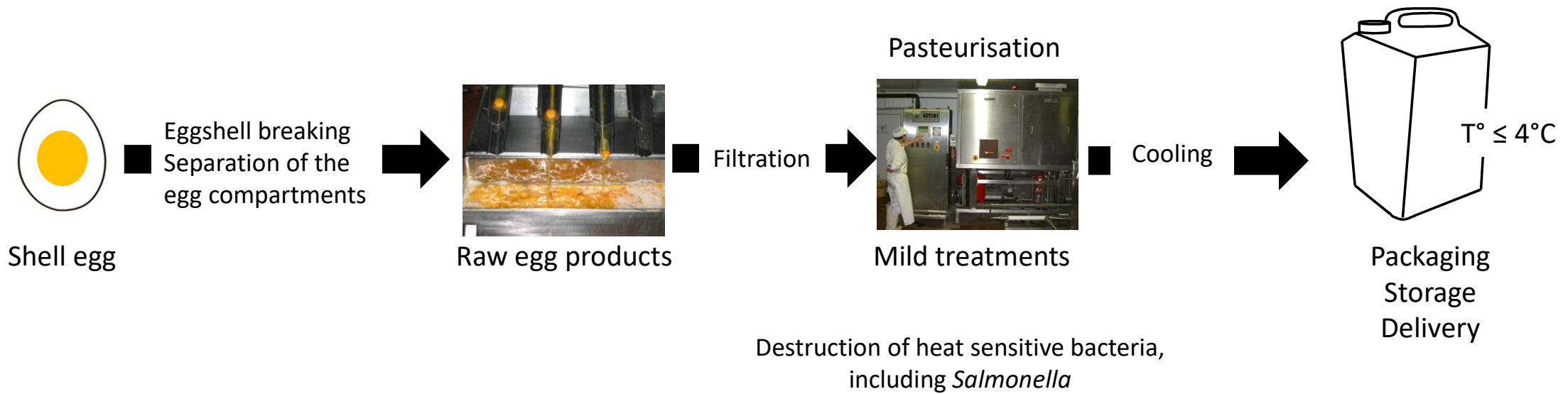
+ Gram-negative bacteria

*Escherichia coli, Yersinia, Enterobacter, Klebsiella, Serratia, Citrobacter, Salmonella, Alcaligenes, Pseudomonas*



Need for egg product stabilization for ensuring egg product safety

# What are the main industrial processes ensuring egg product safety?



What about heat-resistant  
bacteria?

What about  
psychrotrophic  
bacteria?

How to control heat-resistant psychrotrophic bacteria in liquid egg products  
and sensitive food where they are used as ingredients?

# What about the microbiology of egg-based sensitive food?

## Industrial egg-based refrigerated desserts

Sweet egg white foam

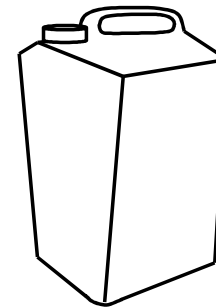
Custard cream comprising whole egg or egg yolk as ingredient



The famous "île flottante" dessert as a research model



?



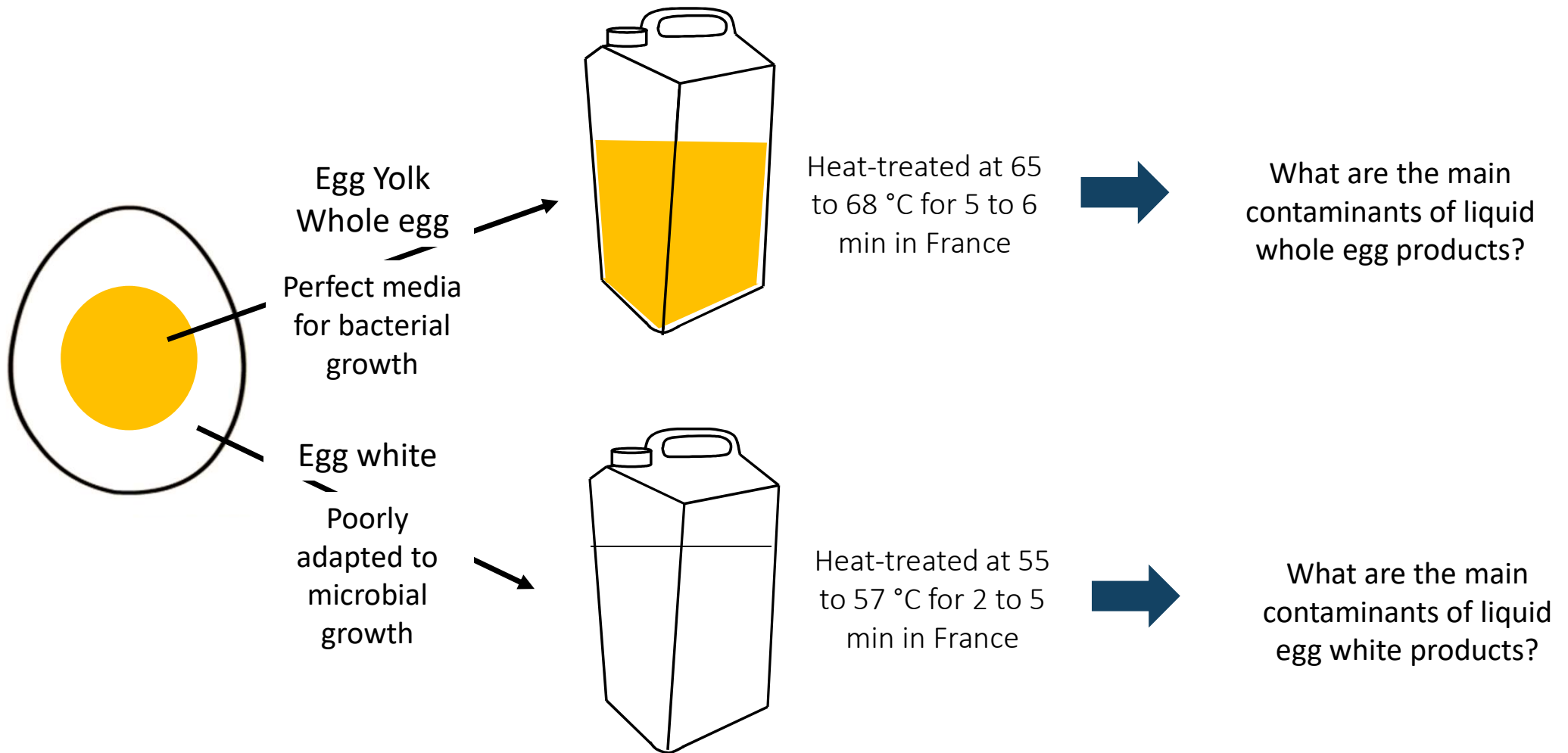
What are the microorganisms coming from egg products ?

How to avoid their presence and/or to control their development ?

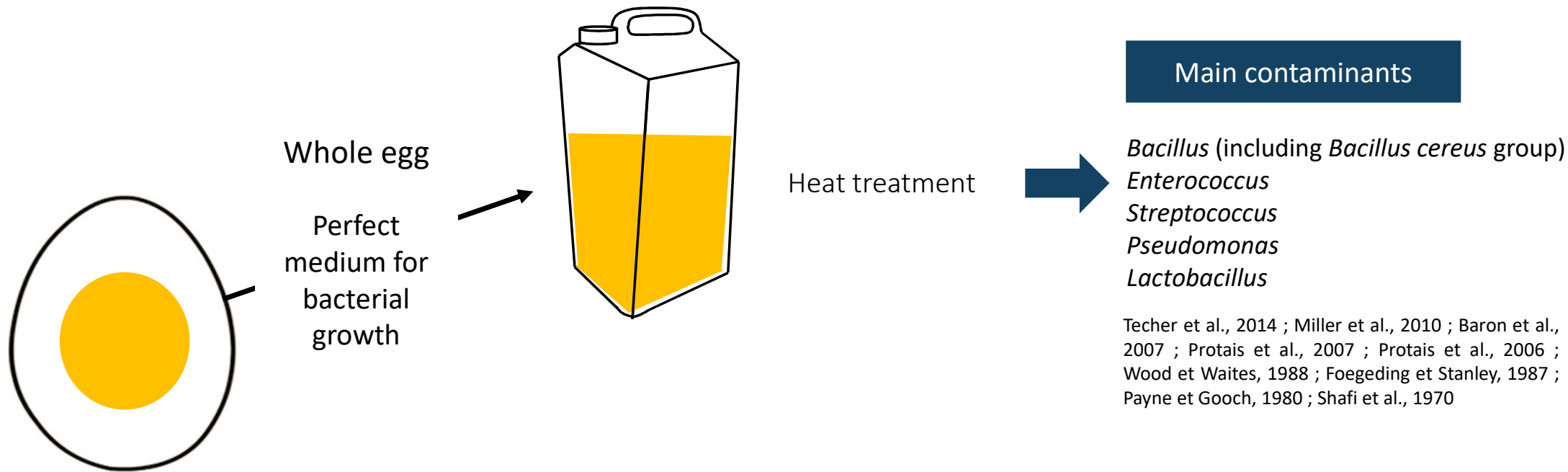


Demand for the development of industrial efficient tools to detect food spoilage contaminants or the products of their metabolism

# What are the main bacterial contaminants of liquid egg products?

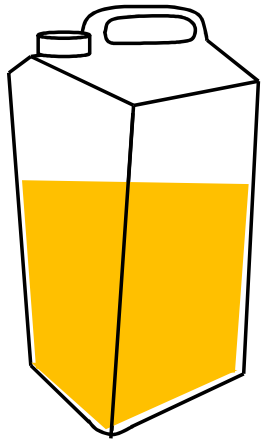


# What are the main bacterial contaminants of liquid egg products?



Techer et al., 2014 ; Miller et al., 2010 ; Baron et al., 2007 ; Protais et al., 2007 ; Protais et al., 2006 ; Wood et Waites, 1988 ; Foegeding et Stanley, 1987 ; Payne et Goch, 1980 ; Shafi et al., 1970

# What are the main bacterial contaminants of liquid whole egg products?



Industrial finished whole egg products coming from 5 French industries

Enrichment of the Gram positive flora

37°C

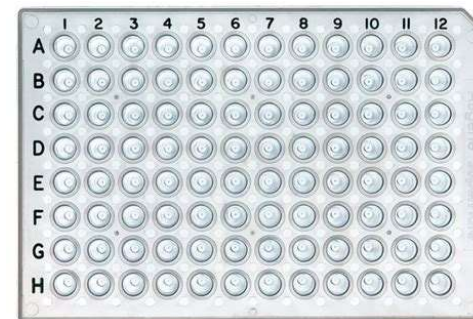
Mesophilic *Bacillus cereus* group bacteria  
100 % of the companies  
76 % of the samples

10°C

Psychrotrophic *Bacillus cereus* group bacteria  
100 % of the companies  
44 % of the samples

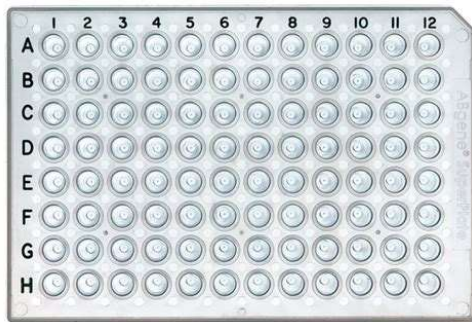
Higher occurrence at the cold season

Koné et al., 2013 BPS

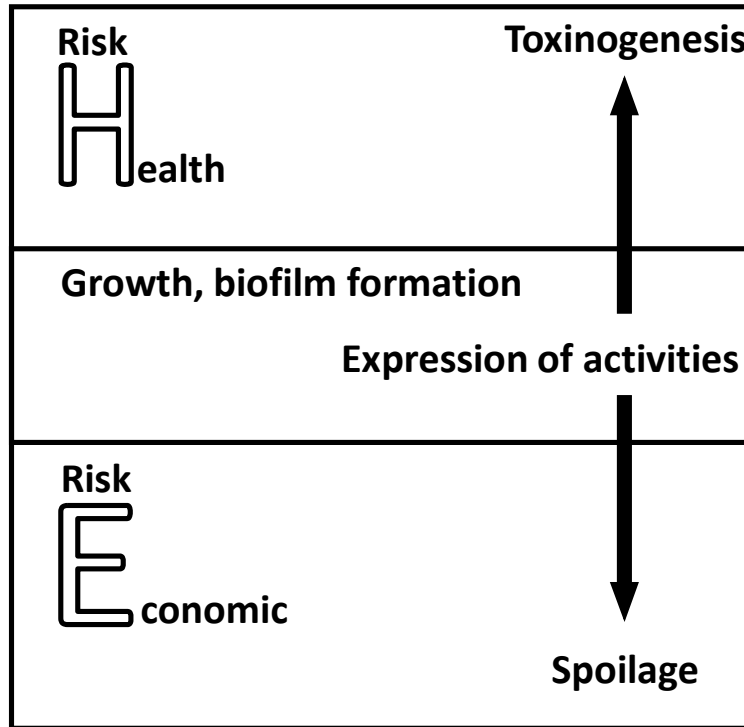


Bacterial collection of 68 representative psychrotrophic *B. cereus* group bacteria

# What are the main features of the bacterial collection coming from whole egg products?



68 psychrotrophic *B. cereus* group bacteria



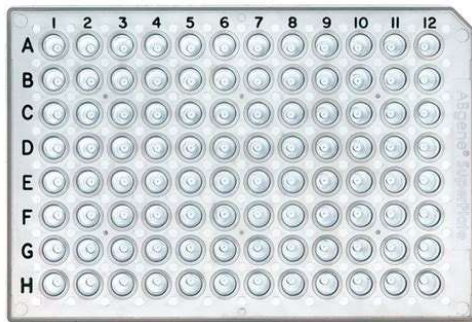
Genetic markers of the psychrotrophy

Belonging to a phylogenetic group (*panC* sequence) (Guinebretière et al., 2008)

Jan et al., 2011. Food Microbiol.

# What are the main features of the bacterial collection coming from whole egg products?

A high diversity inside the collection



68 psychrotrophic *B. cereus* group bacteria



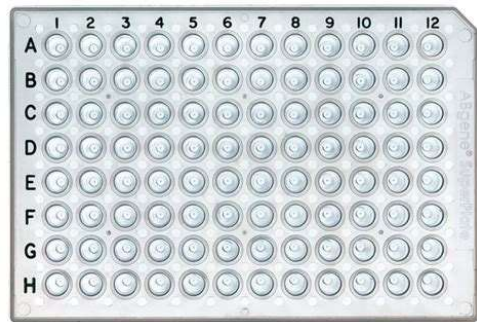
Jan et al., 2011. Food Microbiol.

Belonging to a phylogenetic group (*panC* sequence) (Guinebretière et al., 2008)

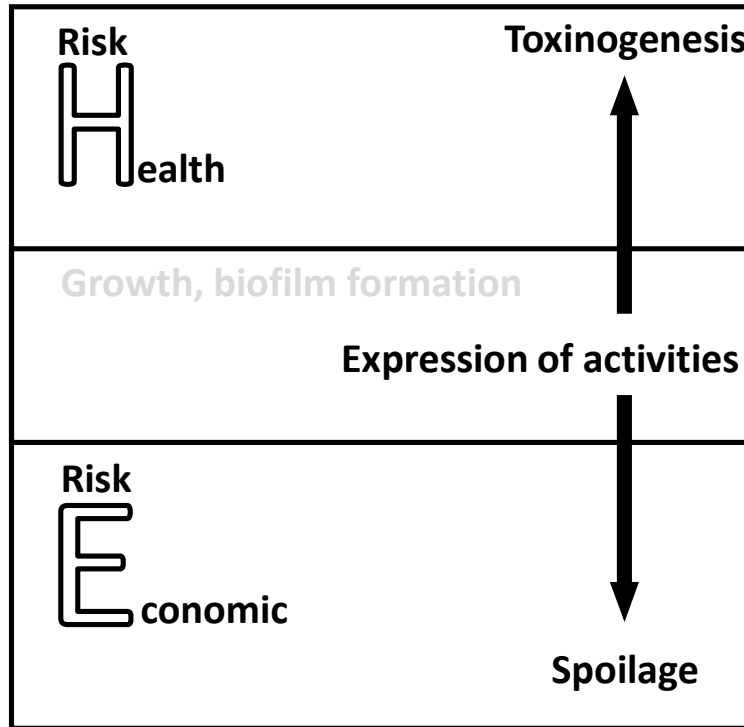
Five phylogenetic groups

Group VI	48.5 %
Group II	41.0 %
Group V	1.5 %
Group IV	7.5 %
Group III	1.5 %

# What are the main features of the bacterial collection coming from whole egg products?

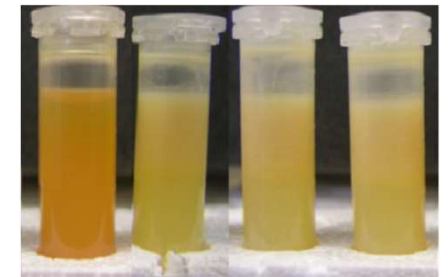
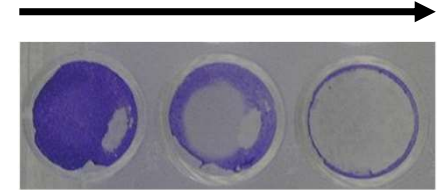


68 psychrotrophic *B. cereus* group bacteria



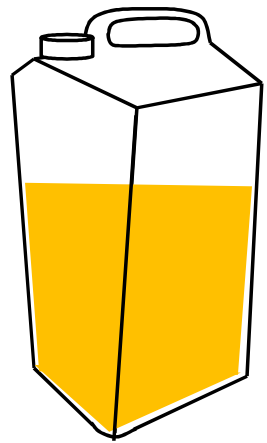
Jan et al., 2011. Food Microbiol.

Cytotoxic activity as a function of time



Spoilage as a function of time

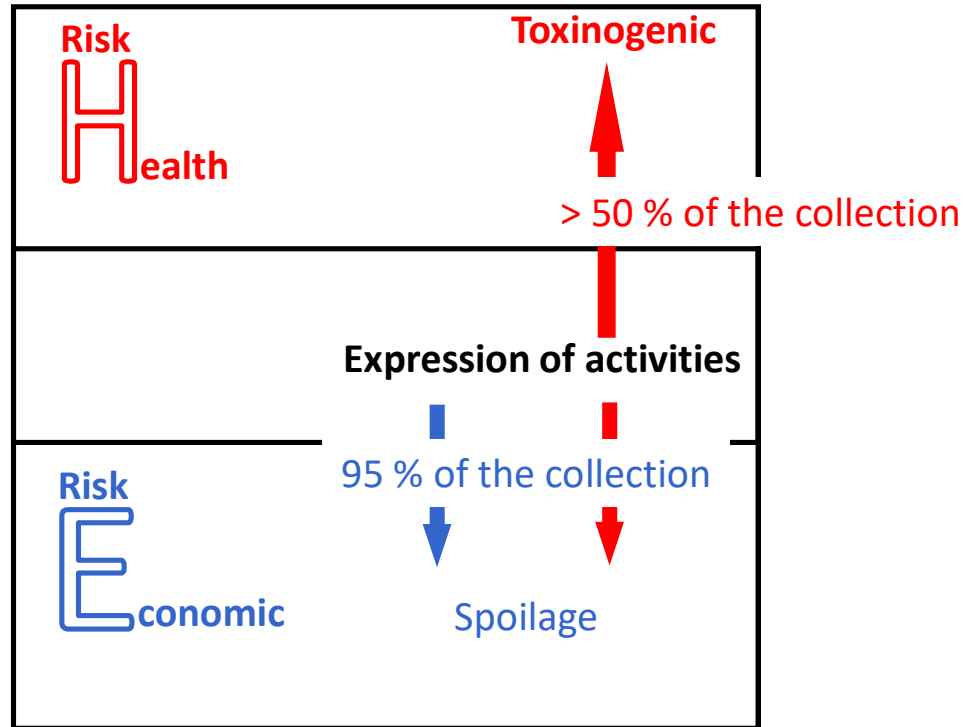
# What are the main features of the bacterial collection coming from whole egg products?



Optimal growth medium

at optimal temperature

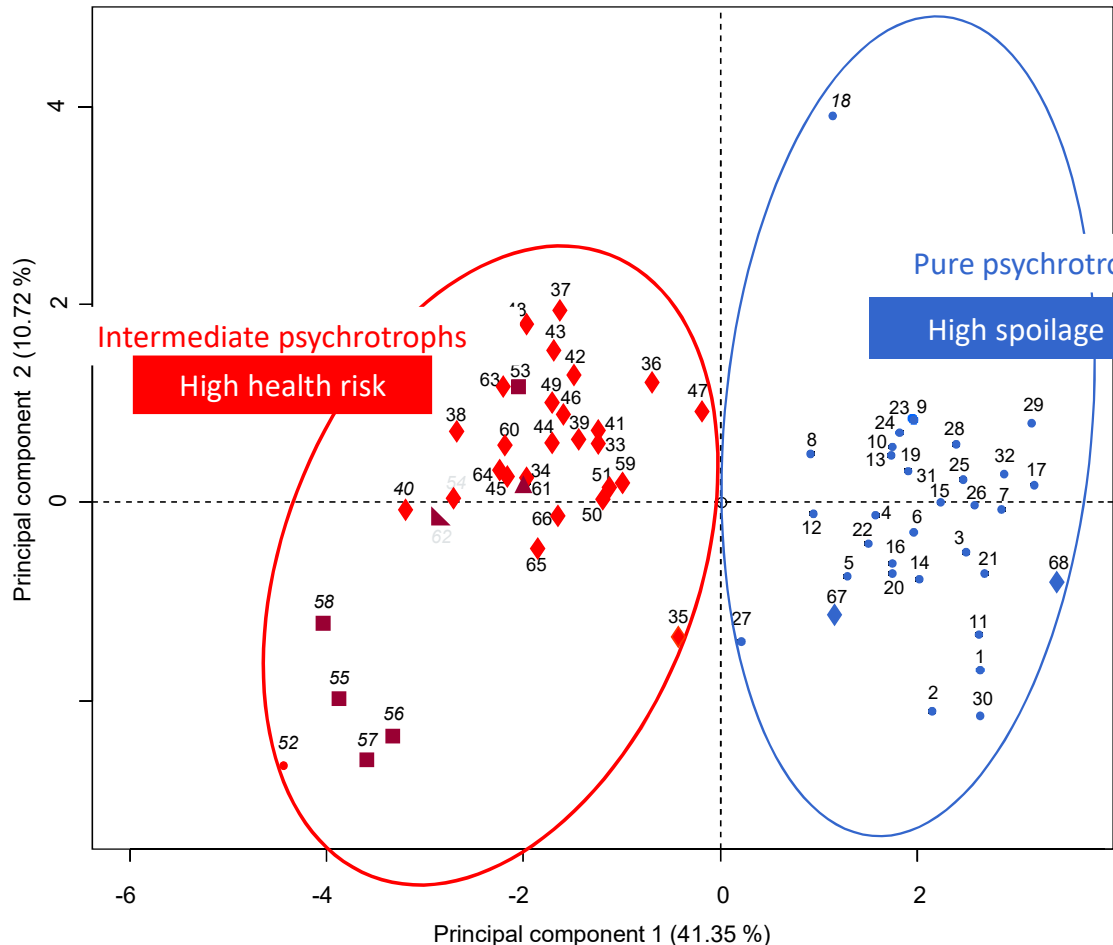
even at low temperature



Jan et al., 2011. Food Microbiol.

# What are the main features of the bacterial collection coming from whole egg products?

An inverse correlation between health and spoilage risks



A psychrotrophic signature in the *cspA* gene

### LEGEND

- ◆ group II
- ▲ group III
- group IV
- ▼ group V
- group VI

In red : isolates associated to health risk

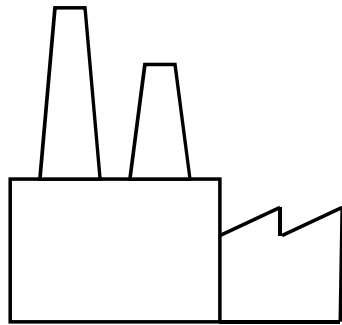
In blue : isolates associated to spoilage risk

In garned red : isolates associated to high sanitary risk

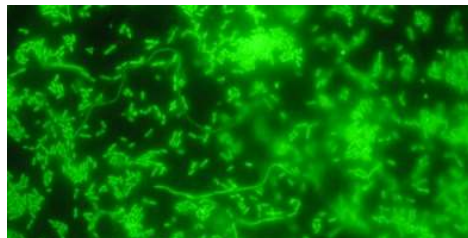
Techer et al., 2014. J. Appl. Microbiol.

# What are the main features of the bacterial collection coming from whole egg products?

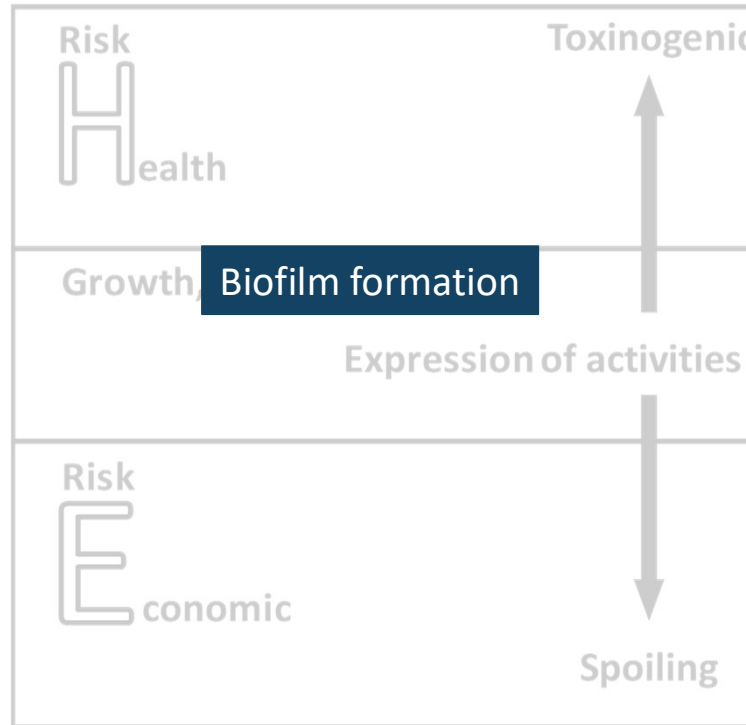
An established risk of biofilm formation



Stainless steel surfaces



**Strong adhesion**

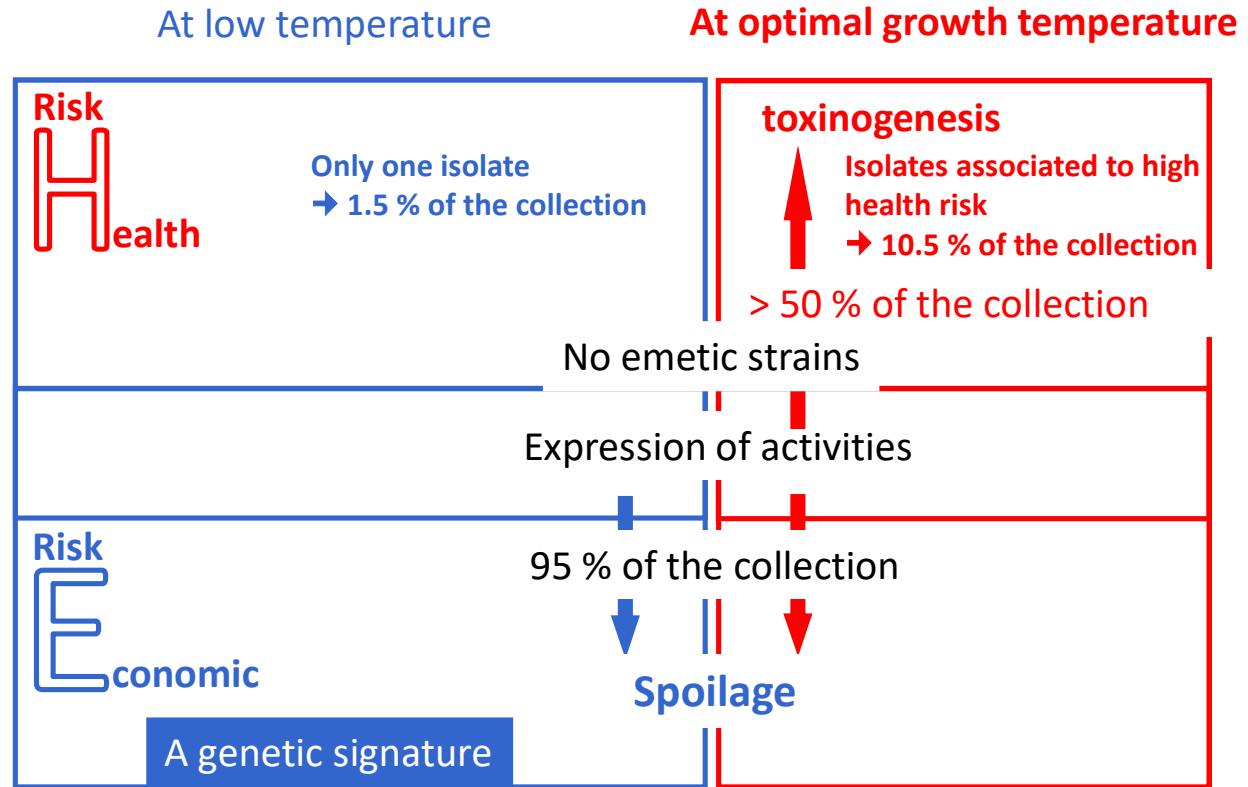
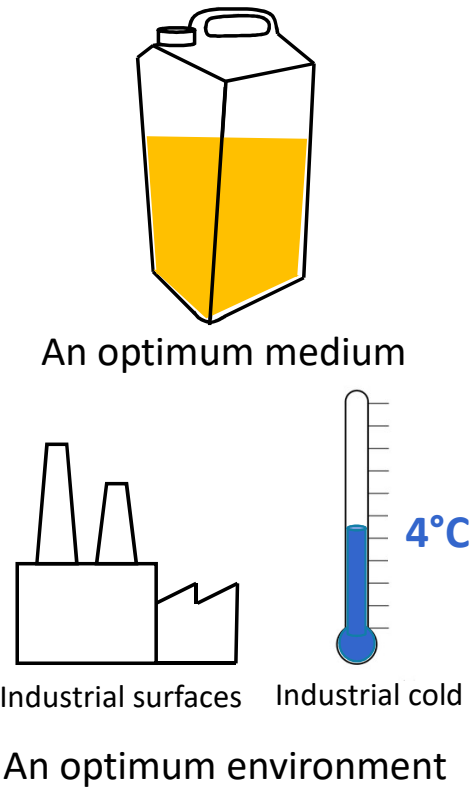


Jan et al., 2011. Food Microbiol.

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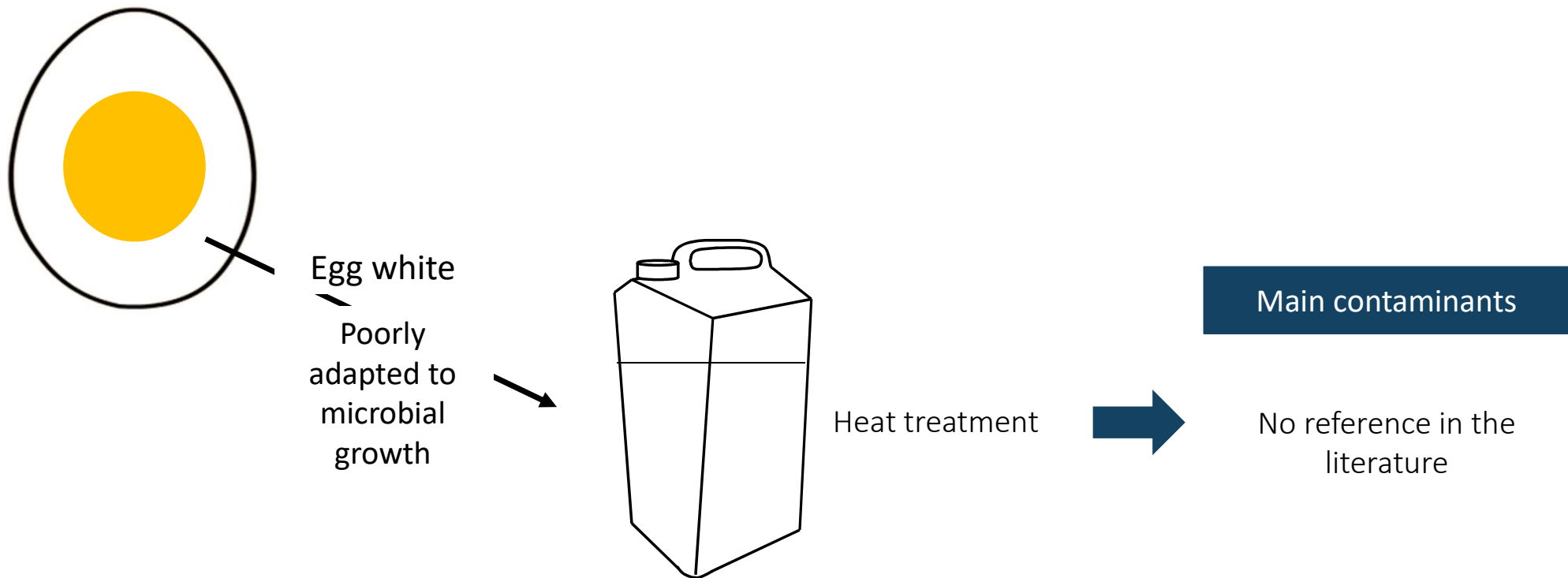
## Selection by the process

## What about the actual risks?

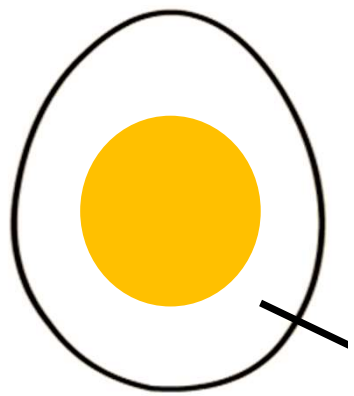


Techer et al., 2014. J. Appl. Microbiol.

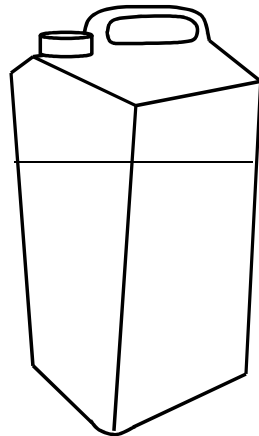
# What are the main bacterial contaminants of liquid egg white products?



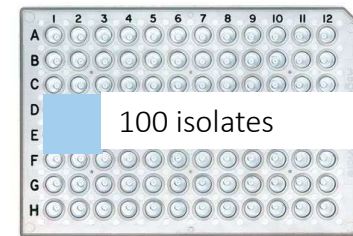
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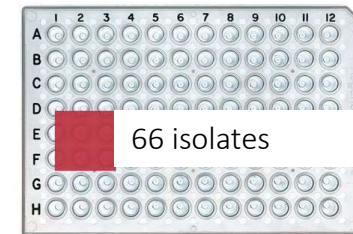
Egg white  
Poorly adapted to microbial growth



Before heat-treatment



After heat-treatment



Techer et al., 2015, J. Food Science

Main contaminants

# What are the main bacterial contaminants of liquid egg white products?

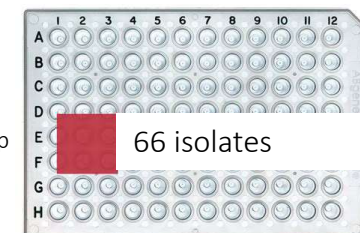
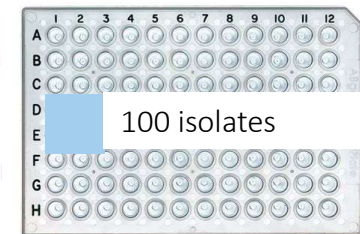
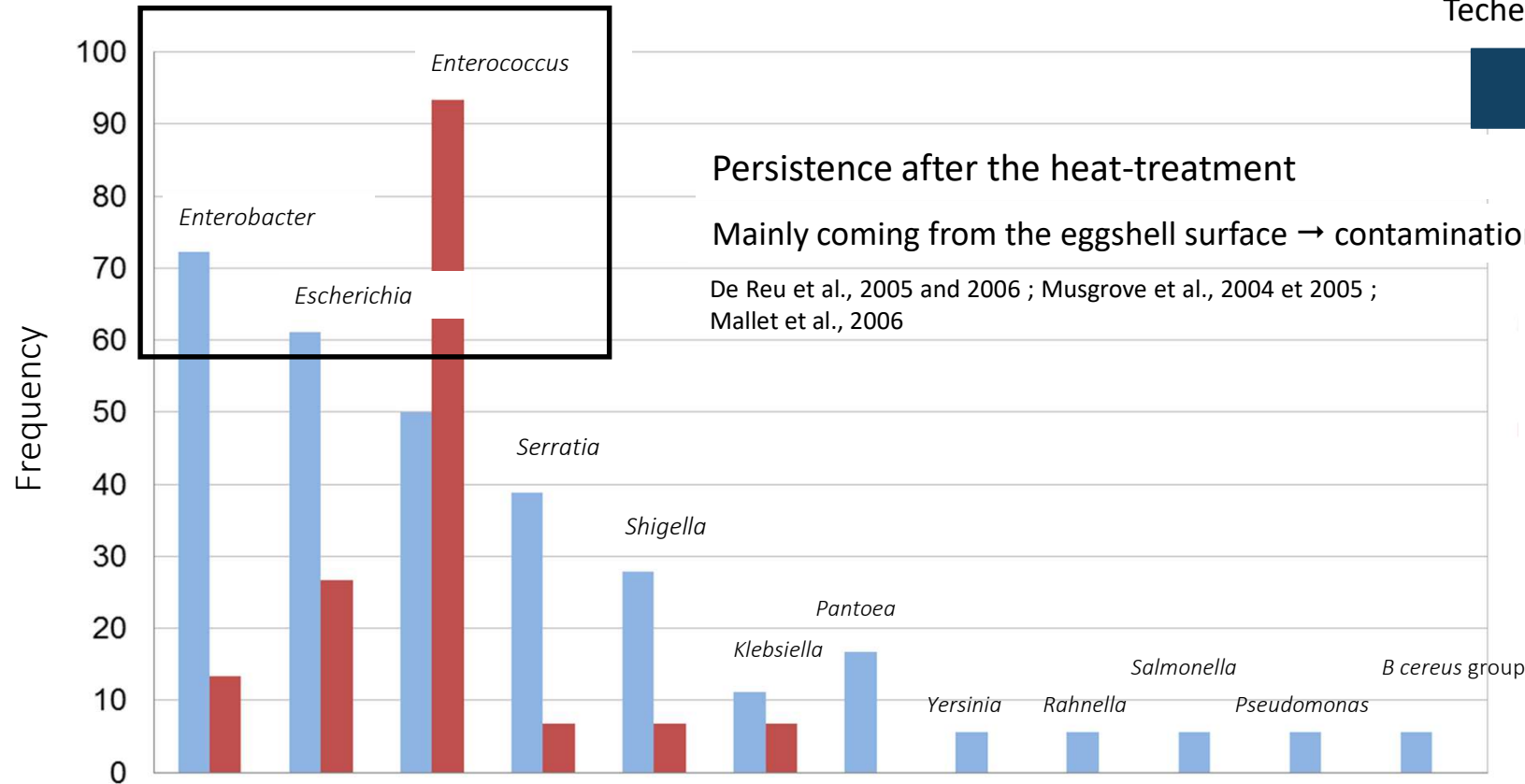
Techer et al., 2015, J. Food Science

## Main contaminants

Persistence after the heat-treatment

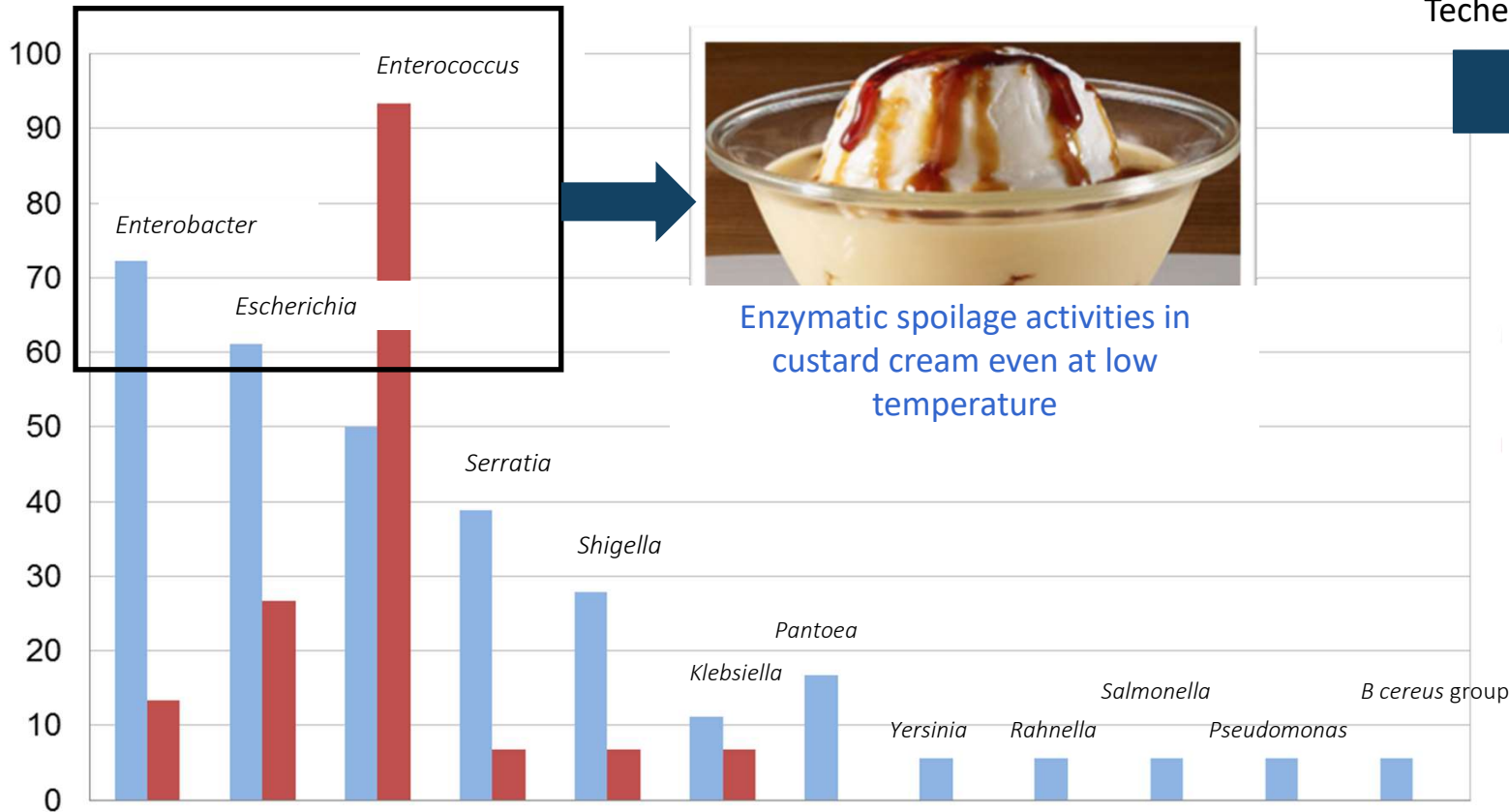
Mainly coming from the eggshell surface → contamination at the egg breeding step

De Reu et al., 2005 and 2006 ; Musgrove et al., 2004 et 2005 ; Mallet et al., 2006



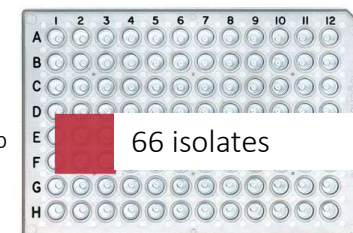
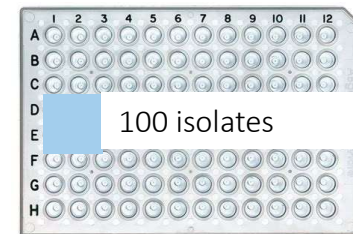
# What are the main bacterial contaminants of liquid egg white products?

Techer et al., 2015, J. Food Science



Enzymatic spoilage activities in custard cream even at low temperature

## Main contaminants



## Conclusion

Persistence of heat-resistant psychrotrophic bacteria in pasteurised liquid egg products  
Spoilage activities, in the product itself and when used as ingredient, even at low temperature

Genetic signature of the spoilage risk due to psychrotrophic *B. cereus* group bacteria

## And now?

Study of the bacterial spoilage metabolism



Development of online tools to early detect food spoilage contaminants or the products of their metabolism



**MERCI**



***THANK YOU FOR  
YOUR ATTENTION***