Policymakers' Event 24.02.2021

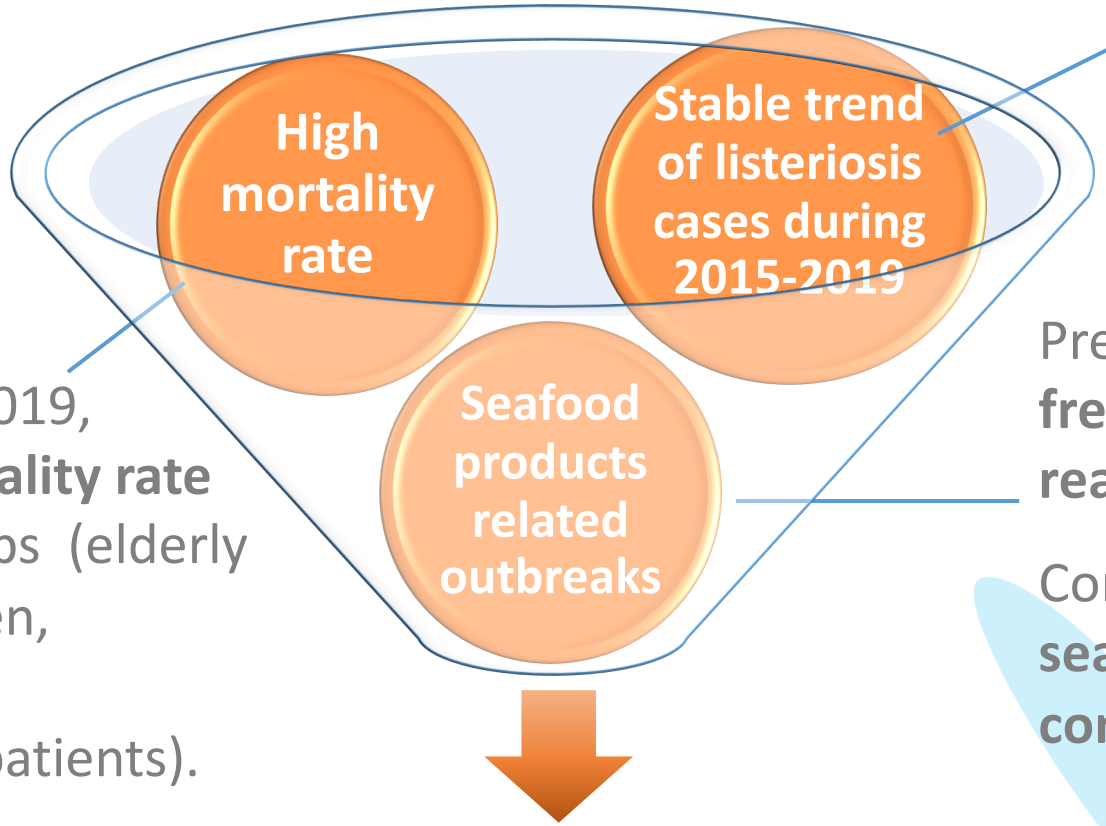
Nutritious, safe, and sustainable seafood for consumers

Listeria-specific bacteriophages for safer ready-to-eat seafood

Amaia Lasagabaster (AZTI)

This project has received funding from the European Union's Horizon 2020 research and innovation programme under Grant Agreement no. 773400 (SEAFOOD TOMORROW). This output reflects the views only of the author(s), and the European Union cannot be held responsible for any use which may be made of the information contained therein.

Listeria monocytogenes responsible for listeriosis



2,621 cases of listeriosis confirmed in **2019.**

300 related deaths in 2019, which implies a high **fatality rate of 17.6%** in at-risk-groups (elderly people, pregnant women, newborns, and immunocompromised patients).

Prevalence of *L.monocytogenes* in **fresh**, **minimally processed** and **ready-to-eat (RTE)** seafood products.

Consumption of contaminated **seafood products** is one of the most **common causes of listeriosis**.

Need to improve the control of *Listeria monocytogenes* in seafood

EFSA and ECDC, 2021

This project has received funding from the European Union's Horizon 2020 research and innovation programme under Grant Agreement no. 773400 (SEAFOOD TOMORROW). This output reflects the views only of the author(s), and the European Union cannot be held responsible for any use which may be made of the information contained therein.

The challenge:
Listeria monocytogenes control

Guidance

Regulation

Regulation (EC) No 2073/2005 of microbiological criteria for food stuffs, including food safety criteria for *Lm*

Seafood Industry
Public Health

Innovative Solutions

Food Safety
Control Strategies

RTE seafood
consumed without further treatment or cooking

This project has received funding from the European Union's Horizon 2020 research and innovation programme under Grant Agreement no. 773400 (SEAFOOD TOMORROW). This output reflects the views only of the author(s), and the European Union cannot be held responsible for any use which may be made of the information contained therein.

Bacteriophages as food safety agents

Bacteriophages (phages) are viruses that **naturally** infect **specific bacteria.**

Each phage can target a **few strains** of a single or different species within the **same genus**

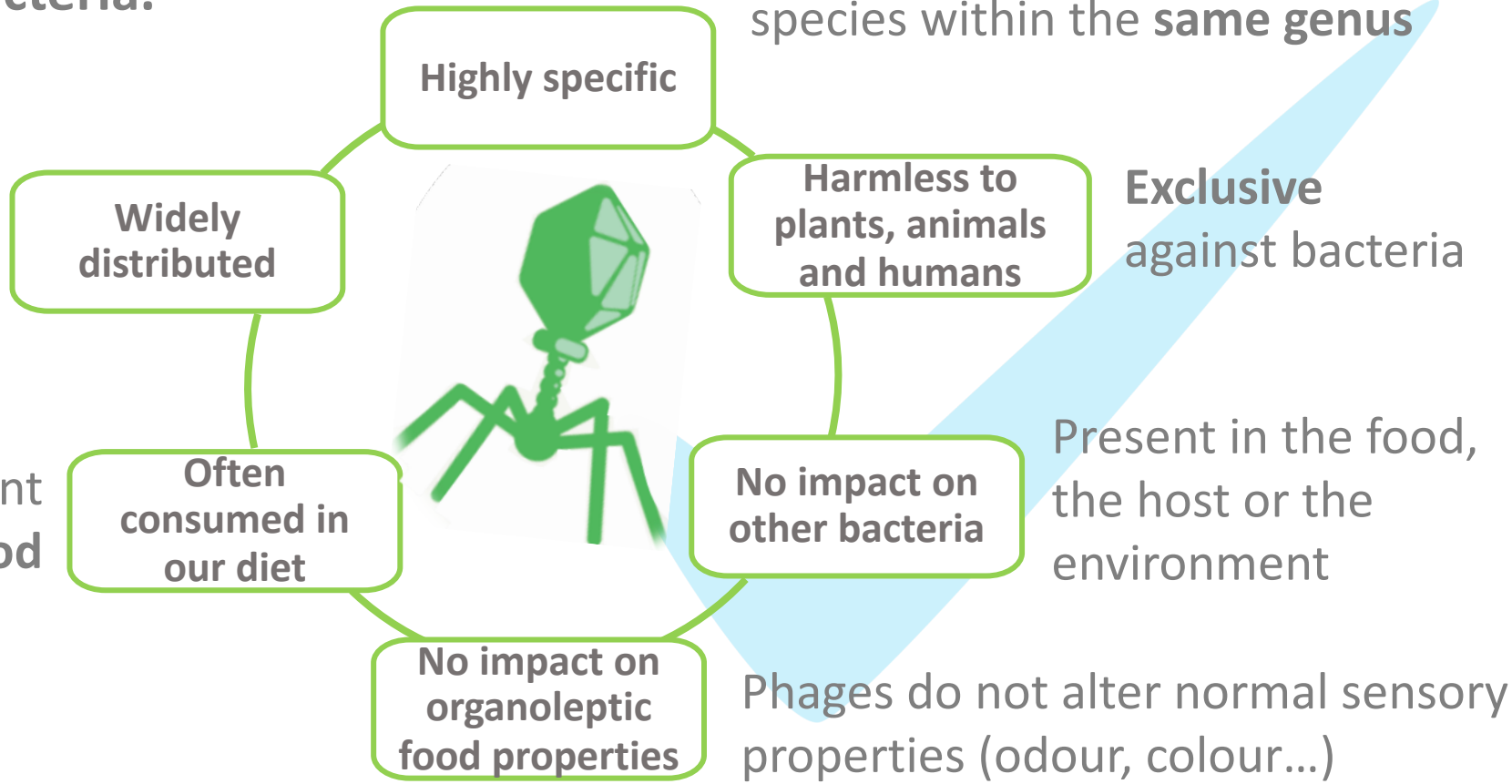
Most abundant organisms on earth: they are **everywhere**

Exclusive against bacteria

Part of the natural microbiota present in the food, including **seafood**

Present in the food, the host or the environment

Phages do not alter normal sensory properties (odour, colour…)



This project has received funding from the European Union's Horizon 2020 research and innovation programme under Grant Agreement no. 773400 (SEAFOOD TOMORROW). This output reflects the views only of the author(s), and the European Union cannot be held responsible for any use which may be made of the information contained therein.

Bacteriophages as food safety agents

Objective

To develop a **bacteriophage-based solution** to control ***L. monocytogenes*** in seafood products

AZTI
MEMBER OF
BASQUE RESEARCH
& TECHNOLOGY ALLIANCE

PTC

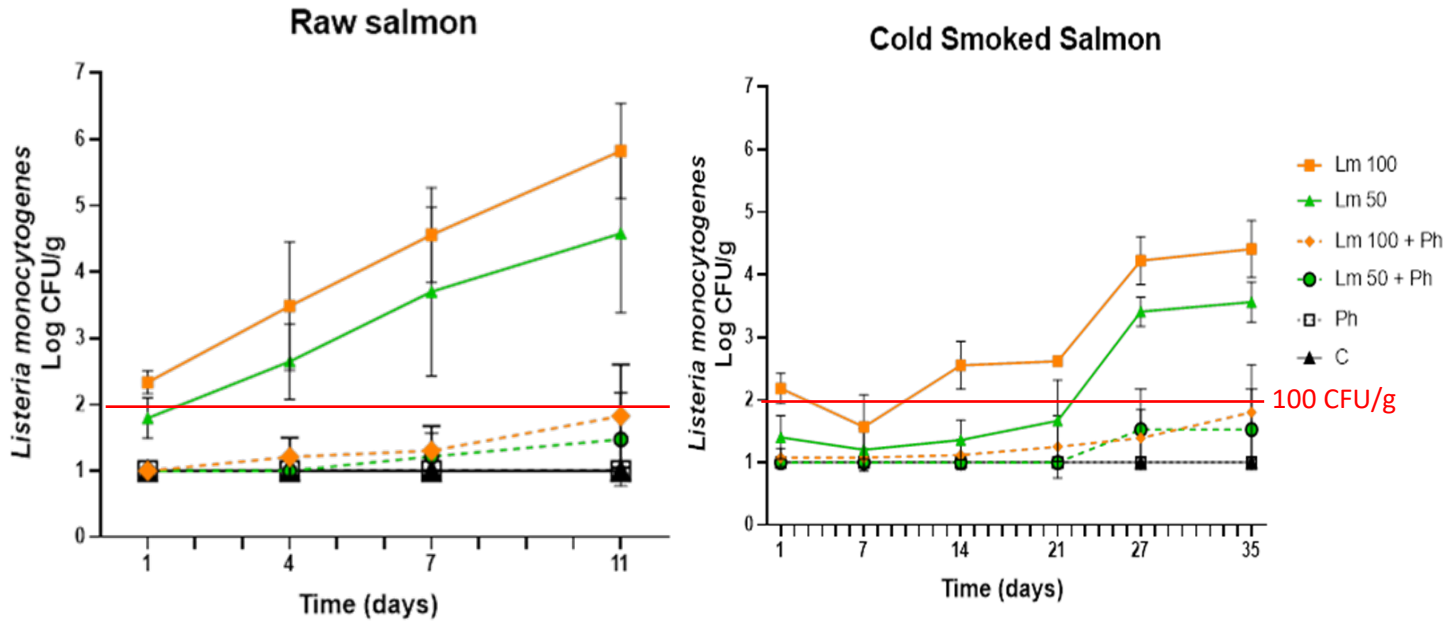
This project has received funding from the European Union's Horizon 2020 research and innovation programme under Grant Agreement no. 773400 (SEAFOOD TOMORROW). This output reflects the views only of the author(s), and the European Union cannot be held responsible for any use which may be made of the information contained therein.

Listeria-specific bacteriophages for safer seafood

- ✓ A *Listeria* specific **bacteriophage cocktail** with 5 well-characterized **complementary phages**
- ✓ Effective against a **broader range** of *Listeria* strains than single monophages
- ✓ **> 90% of *Listeria* isolates** of relevance for the seafood industry
- ✓ **Stable** under application conditions of **temperature (4-25˚C)** and **pH (2-9.5)**
- ✓ Shelf life of **6 months**
- ✓ Able to be **produced at industrial commercial scale** to optimal yields

This project has received funding from the European Union's Horizon 2020 research and innovation programme under Grant Agreement no. 773400 (SEAFOOD TOMORROW). This output reflects the views only of the author(s), and the European Union cannot be held responsible for any use which may be made of the information contained therein.

Listeria-specific bacteriophages for safer seafood



- ✓ Able to **reduce** initial ***L.monocytogenes*** **load** and keep it **below the legal limit** of 100 CFU/g at the end of the shelf-life of both raw salmon and cold smoked salmon.

This project has received funding from the European Union's Horizon 2020 research and innovation programme under Grant Agreement no. 773400 (SEAFOOD TOMORROW). This output reflects the views only of the author(s), and the European Union cannot be held responsible for any use which may be made of the information contained therein.

Regulatory status

Only few countries have developed legislation concerning food applications of bacteriophages:

- **USA**: USDA approval as **processing aid** and **GRAS-status** by FDA after showing that application is safe and provide a benefit for the consumer.
- **Switzerland**, **Israel**, **Canada**, **Australia**, **New Zealand** or **Brazil:** Health authorities approved specific applications as **processing aid.**
- **EU: No clear path** on how to register a phage product.
  - Three positive scientific opinions of the EFSA Panel on Biological Hazards (BIOHAZ): safe and effective.
  - European Commission did not agree on a regulatory path on the use of phages in foods, and relegated the responsibility back to the individual member states.
  - Discrepancies about whether phages should be considered as food aditives (labelled) or as processing aids.
  - **The Netherlands** approved a **temporary authorization** for the application of phages as **processing aid** in chicken meat.

efsa
European Food Safety Authority

EFSA Journal 2012;10(3):2615

SCIENTIFIC OPINION

Scientific Opinion on the evaluation of the safety and efficacy of Listex™ P100 for the removal of *Listeria monocytogenes* surface contamination of raw fish[1]

EFSA Panel on Biological Hazards (BIOHAZ)[2, 3]

European Food Safety Authority (EFSA), Parma, Italy

This scientific output, published on 19 April 2012, replaces the earlier version published on 20 March 2012.*

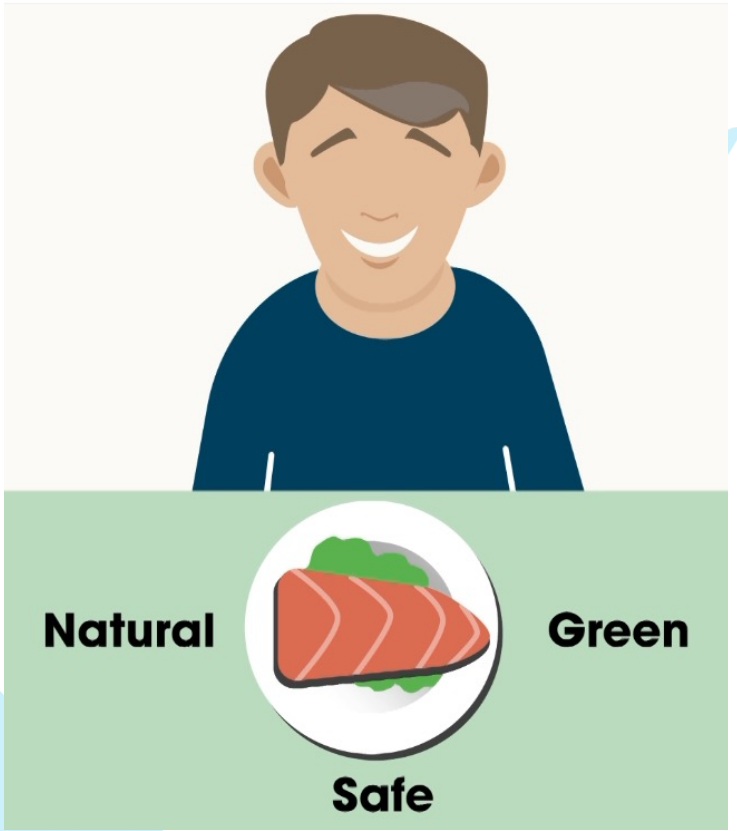
ABSTRACT

Studies evaluating the safety and efficacy of Listex™ P100 to reduce *Listeria monocytogenes* contamination on raw fish were assessed. The material should not present human toxicological problems because the bacteriophage P100, used as active principle, is not regarded as harmful to consumers nor to organisms other than *Listeria* spp., and because the fabrication parameters do not include anything obvious that might compromise safety. Data of studies considered indicate that Listex™ P100 is listericidal on inoculated catfish and salmon samples, but do not allow definitive conclusions on efficacy in reducing *L. monocytogenes* counts on raw fish nor on its impact on *L. monocytogenes* contamination levels in finished product. It was not possible to estimate the potential listeriosis risk reduction by treating raw fish with Listex™ P100. The data were not adequate to allow firm conclusions on persistence or activity of P100 in stored fish. The proposed use of Listex™ P100 is unlikely to result in emergence of reduced susceptibility to biocides and/or resistance to key therapeutic antimicrobials: however, this conclusion may need verification. No information was provided on survival of P100 in processing wastewater or the environment, or on the potential accumulation of naturally P100 resistant *L. monocytogenes* variants. Pilot and industrial scale studies should consider parameters affecting decontaminating efficacy, and should verify that application on raw fish has an impact on reduction of *L. monocytogenes* contamination on the final product. The persistence or activity of P100 as well as potential changes in *L. monocytogenes* counts should be evaluated during fish storage. Tests to investigate potential development of resistance or reduced susceptibility to biocides and key therapeutic

This project has received funding from the European Union's Horizon 2020 research and innovation programme under Grant Agreement no. 773400 (SEAFOOD TOMORROW). This output reflects the views only of the author(s), and the European Union cannot be held responsible for any use which may be made of the information contained therein.

Conclusions

- ✓ Biological properties of lytic bacteriophages, as well as the new data on the effectiveness of *Listeria*-specific bacteriophages on seafood products, make **bacteriophage biocontrol** a promising strategy for **seafood safety applications**.
- ✓ Use of ***Listeria* bacteriophages** could contribute as an **additional innovative solution** in a multi-hurdle approach to control the prevalence of *L. monocytogenes* in seafood products.



This project has received funding from the European Union's Horizon 2020 research and innovation programme under Grant Agreement no. 773400 (SEAFOOD TOMORROW). This output reflects the views only of the author(s), and the European Union cannot be held responsible for any use which may be made of the information contained therein.

Thank You

Contact Details:

Amaia Lasagabaster alasa@azti.es

@SEAFOOD_TMRW

This project has received funding from the European Union's Horizon 2020 research and innovation programme under Grant Agreement no. 773400 (SEAFOOD TOMORROW). This output reflects the views only of the author(s), and the European Union cannot be held responsible for any use which may be made of the information contained therein.