**Sara Bover-Cid**

Food Safety Programme
Food Industry Area

# Outsourcing activity 1: literature search on *L. monocytogenes* in a wide range of RTE foods

Stakeholder meeting on draft scientific opinion on *Listeria monocytogenes* contamination of ready-to-eat foods and the risk for human health in the EU

Parma, 19-20 September 2017

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# Outline of the presentation

I. The consortium
II. Objectives and methodology
III. Results and outputs
IV. Conclusions

# I. The Consortium

Closing gaps for performing a risk assessment on *Listeria monocytogenes* in ready-to-eat (RTE) foods: Activity 1, an extensive literature search and study selection with data extraction on *L. monocytogenes* in a wide range of RTE food. Contract number: NP/EFSA/BIOCONTAM/2015/04-CT1

**IRTA** is a research institute owned by the Government of Catalonia ascribed to the Department of Agriculture. IRTA researchers have a large expertise in food microbiology, particularly focused on food-oriented and applied approach, including predictive microbiology. The research group has extensive know-how about *Listeria monocytogenes* in food endorsed by a number of national and international projects, scientific publications and communications. Participated in the EFSA contract (OC/EFSA/BIOCONTAM/2014/02CT1, Activity 2).

**HIBRO** is a research group belonging to the Depart of Food Science and Technology at University of Córdoba (**UCO**, Spain). HIBRO activities are focused on Food Safety and Quality aspects with an extensive mathematical background and an internationally recognized expertise in the development of "Predictive Microbiology" models and Quantitative Microbial Risk Assessment (QMRA) studies for different foodborne pathogens such as *Listeria monocytogenes*. Coordinated the EFSA contract (OC/EFSA/BIOCONTAM/2014/02CT1, Activity 2).

# II. Objectives and methodology

To perform an extensive literature search aiming:

1. to describe the **occurrence and levels of contamination** of *L. monocytogenes* in ready-to-eat (RTE) foods;
2. to describe the **risk factors** of the *L. monocytogenes* contamination in different RTE foods.

***review question 1***
PO

***review question 2***
PECO

Scope

hot or cold smoked or gravad fish, ready-to-eat meat products, cheeses, retail unpasteurized milk, melons and leafy greens **available at processing or later stages** (pre-harvest products excluded).

- Time span: 1990- to present
- The quality appraisal of the selected studies was out of the scope of the activity.


EFSA (2010)

# II. Objectives and methodology

Specific objectives

1 to update **literature searches** (search protocol provided)

Search strings (Appendix A)

Databases: SCI-EXPANDED
MEDLINE

Grey literature

THOMSON REUTERS
Web of Science

EN

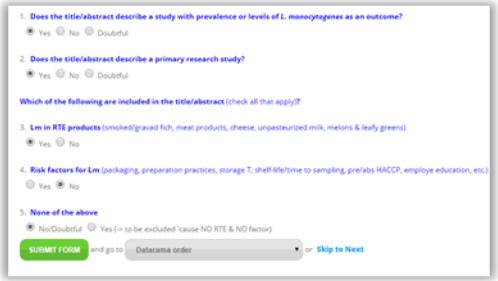
2 to perform a **study selection** at:

- Level 1 for relevance
- Level 2 for eligibility

Title & abstract (provided criteria)

Full text (criteria agreed with EFSA)

DistillerSR

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# Level 2 screening for eligibility

## Selection/exclusion criteria

**Proposed eligibility and exclusion criteria for the review question 2:**
**to describe the risk factors of the *L. monocytogenes* contamination in different RTE foods.**

| Key element | Eligibility criteria | Exclusion criteria |
|---|---|---|
| **Population** | • **RTE food** (i.e. in the definition provided by Regulation (CE) 2073/2005), e.g. these included in EU-wide BLS (cooked meat products, smoked and gravad fish, cheese), unpasteurized milk, melons and leafy greens. Other such as fruits, cured/fermented meat products | • Non-RTE food (i.e. out of the definition provided by Regulation (CE) 2073/2005). |
| | • Samples **collected at processing or later** (retail phase) from European countries | • Pre-harvest food samples |
| | • Samples manufactured by food processors (**commercial** products) | • Food samples manufactured *ad-hoc* for research experiments |
| **Outcome** | • Occurrence/prevalence and/or levels (in terms of concentration) of ***L. monocytogenes*** | • Data about other *Listeria* species and *Listeria* spp. |
| **Exposure and comparator** | • **Risk factors** associated with:<br>○ processing **environment** (e.g. presence/absence of HACCP system, education and training of food handlers, validated cleaning and disinfection programme, food-contact surface testing/results)<br>○ **manufacturing** and preparation practices (e.g. type of processing, exposure after a lethal treatment, for instance during slicing and packaging, use of post-lethally treatment and/or antimicrobial process)<br>○ **product** characteristics (e.g. pH, $a_w$, salt, preservatives, packaging type)<br>○ **storage** conditions (e.g. time and temperature) | |
| **Study design** | • Survey studies about **naturally exposed** products | • Challenge test, i.e. dealing with deliberately inoculated products |
| | • Studies evaluating *L. monocytogenes* control strategies (e.g. cleaning and disinfection, post-processing listericidal treatments, growth inhibitors) in non-inoculated products | • Intervention experimental studies, samples from batches with the intervention and inoculated samples |
| | • Outbreak investigations and recalls when risk factors associated with the pathogen levels in incriminated food are reported | • Studies evaluating the performance/ accuracy of analytical methodologies in deliberately inoculated samples |
| **Type of publication** | • **Primary research** study | • Review articles, editorials and letters to editor |
| **Language restriction** | • English, Spanish, French, Portuguese or Italian | • Any other language |

# II. Objectives and methodology

Specific objectives

1 to update **literature searches** (search protocol provided)

Search strings (Appendix A)

Databases: SCI-EXPANDED
MEDLINE

Grey literature

THOMSON REUTERS
Web of Science

EN

2 to perform a **study selection** at:

- Level 1 for relevance
- Level 2 for eligibility

Title & abstract (provided criteria)

Full text (criteria agreed with EFSA)

DistillerSR

(Appendix B)

3 to **extract data** and create evidence table

(1) General information about the study
(2) RTE food sample and analytical procedure
(3) Risk factors in relation to RQ 2
(4) Outcome (Lm prevalence and conc)

4 to **synthetize data** to reach the general objectives

External Scientific Report & suppl

EFSA Supporting Publications Open Access WILEY

(Appendix C&D)

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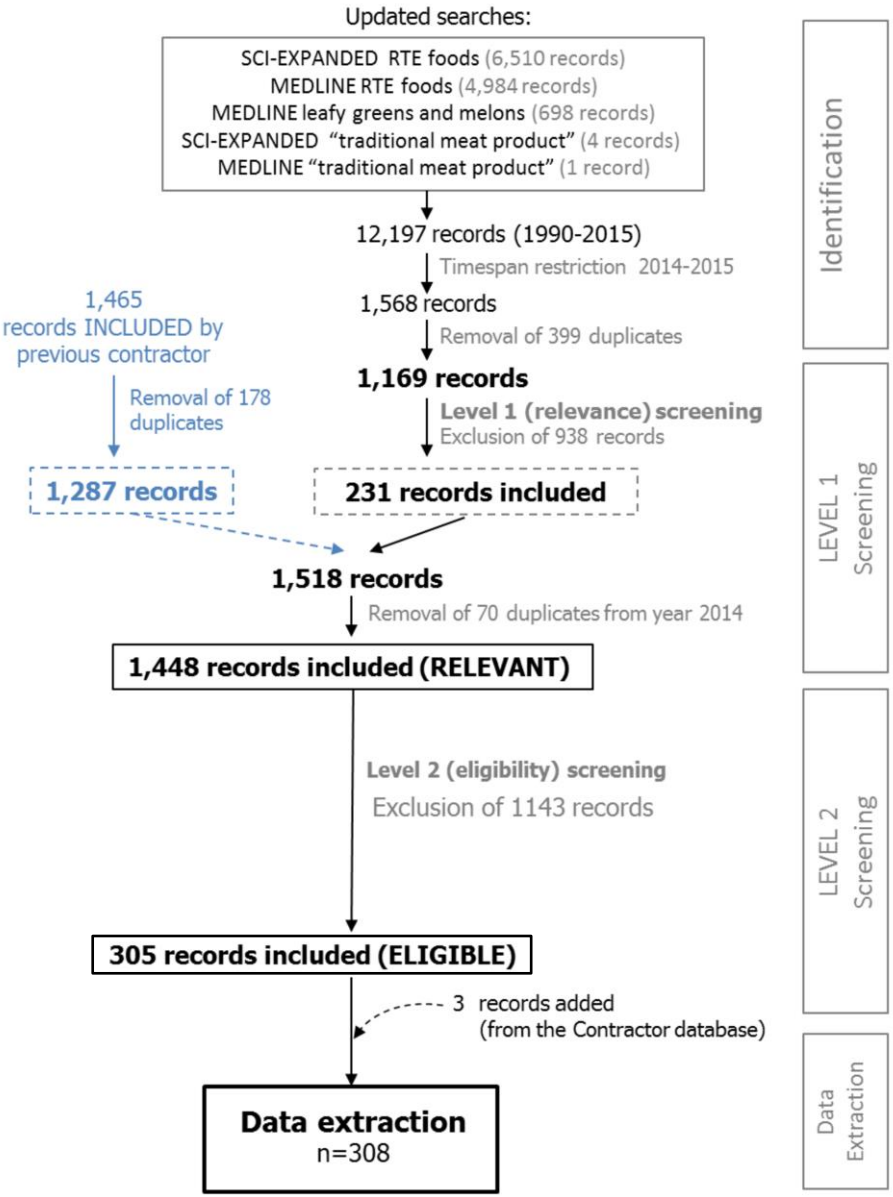
# III. Results and Outputs

## Specific objectives

1 to update **literature searches** (search protocol provided)

2 to perform a **study selection** at:

- Level 1 for relevance
- Level 2 for eligibility




*Flowchart showing the process and results of record identification (search), level 1 screening of records (in updated search) for relevance, merging of libraries and level 2 screening for eligibility and data extraction*

# III. Results and Outputs

3 to **extract data** and create evidence table

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# III. Results and Outputs

Data extraction (***outcome***)

**Table D. 9:** Prevalence and levels of *Listeria monocytogenes* in meat products

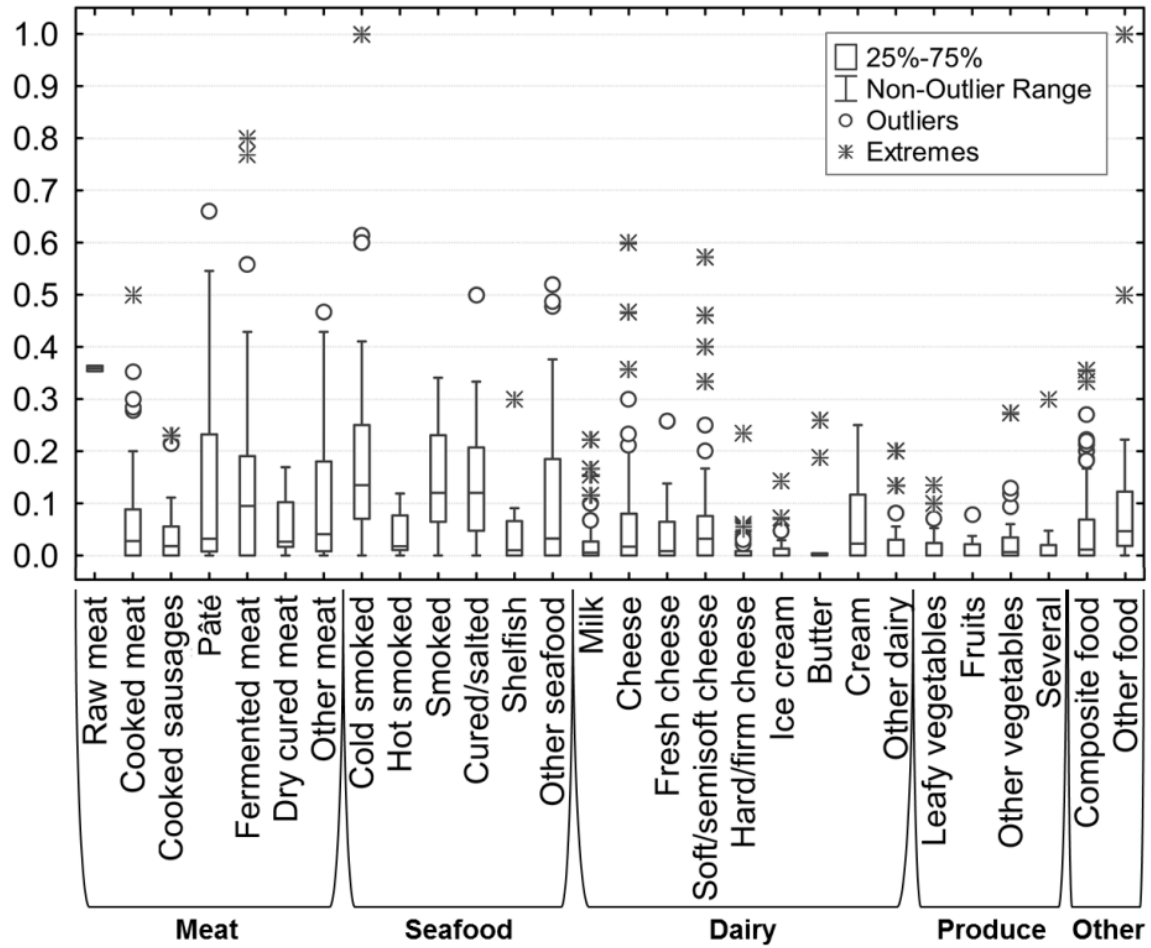
| | | | | | | Semi-quantitative levels | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **RefID[a]** | **Product** | **n** | **s** | **Prevalence** | **Levels** | **>10-100** | **>100-1,000** | **>1,000-10,000** | **>10,000-100,000** | **>100,000-1,000,000** | **>10** | **<10** | **Other[c] (level,n)** | **Units** | **% >100** |
| | **Cooked meat** | | | | | | | | | | | | | | |
| **255** | Cooked ham | 487 | 8 | 0.016 | NR | | | | | | | | | | |
| **665** | Vacuum packaged cooked sliced meat (after slicing and packaging) | 127 | NR | | SQ | 0 | | | | | | | | CFU/g | 0 |
| **665** | Vacuum packaged cooked sliced meat (end of shelf-life) | 127 | NR | | SQ | 10 | | | | | | | | CFU/g | 0 |
| **1182** | Cooked meat products (in-store packaged) | 369 | 34 | 0.092 | NR | | | | | | | | | | |
| **1206** | Heat-treated meat products | 14 | 0 | 0.000 | NR | | | | | | | | | | |
| **1208** | Precut (sliced or cubed) RTE heat-treated meat products | 160 | 13 | 0.081 | NR | | | | | | | | | | |
| **1223** | Cooked ham | 67 | 0 | 0.000 | NR | | | | | | | | | | |
| **1326** | Cooked ham | 24 | 3 | 0.125 | SQ | | | | | | | | | | 0 |
| **1326** | Cooked turkey breast & Pork luncheon meat | 24 | 4 | 0.167 | SQ | | | | | | | | | | 0 |
| **1434** | RTE meat | 18 | 5 | 0.278 | NR | | | | | | | | | | |
| **1481** | Cold meats | 2078 | 60 | 0.029 | SQ | | | | | | | | | | 0.03 |
| **1538** | Deli meat products (Vacuum packaged by producer) | 220 | 6 | 0.027 | SQ | 0 | 2 | | | | | 4 | <10,4 | CFU/g | 0.9 |
| **1538** | Deli meat products (in-store packaged) | 200 | 17 | 0.085 | SQ | 3 | 7 | | | | | 6 | >1000,1 | CFU/g | 4 |
| **1563** | Cold, sliced, RTE meats (cut/slied on or off the premises) | 3455 | NR | | SQ | | 5 | 0 | | | | | <20,3442; <100,8 | CFU/g | 0.14 |
| **1729** | RTE pre-cooked chilled chicken | 102 | 29 | 0.284 | NR | | | | | | | | | | |
| **1742** | Heat treated meat products (brawn, liver sausages, hamburger, susages, ham) | 112 | 17 | 0.152 | NR | | | | | | | | | | |

# III. Results and Outputs

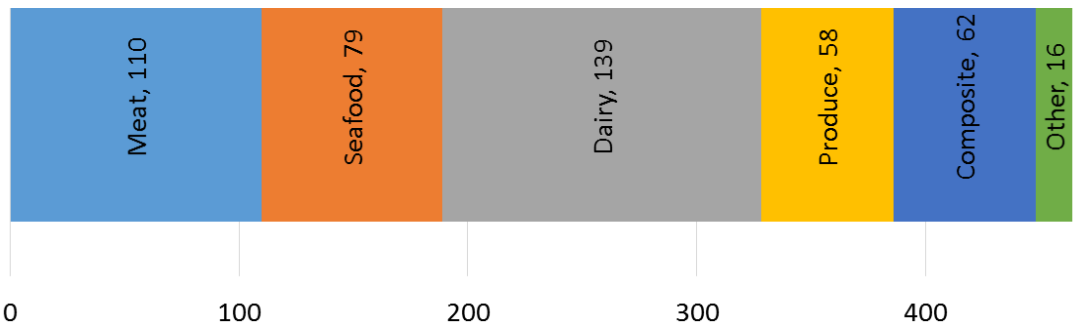


Box-plot of the **prevalence** data extracted by ready-to-eat (RTE) food subcategory

*Median value is indicated in the interquartile box. Outliers (O) and extreme (∗) values correspond to values at 1.5 and 3-fold the interquartile range.*




Distribution of the RTE food products included in the studies of the eligible records and grouped in the pre-defined food categories



# III. Results and Outputs

Data extraction (***outcome vs risk factors***)

**Table 8:** Prevalence data of *Listeria monocytogenes* contamination of RTE food recorded in intervention studies

| RefID | Product | N | S | Prevalence (S/N) |
|---|---|---|---|---|
| **1922** | Vacuum-packaged cold-smoked salmon (time 0) | 360 | 26 | 0.072 |
| | m-packaged cold-smoked salmon (not superchilled) | 198 | 51 | 0.258 |
| | m-packaged cold-smoked salmon (superchilled 14 days) | 132 | 33 | 0.250 |
| | m-packaged cold-smoked salmon (superchilled 28 days) | 132 | 30 | 0.227 |
| **1952** | Pâté (slices from loaves on display) | 155 | 46 | 0.297 |
| | Pâté (unopened packs) | 50 | 23 | 0.460 |
| | Pâté (vacuum-packaged portions) | 11 | 6 | 0.545 |
| | Pâté (7 loaves of 2 kg, 21 days storage at 4ºC) | 56 | 37 | 0.661 |
| **1231** | Cold-smoked rainbow trout (before eradication programme) | 22 | 22 | 1.000 |
| | Cold-smoked rainbow trout (after eradication programme) | 22 | 0 | 0.000 |

N: number of analysed samples; RTE: ready-to-eat; S: number of positive samples.

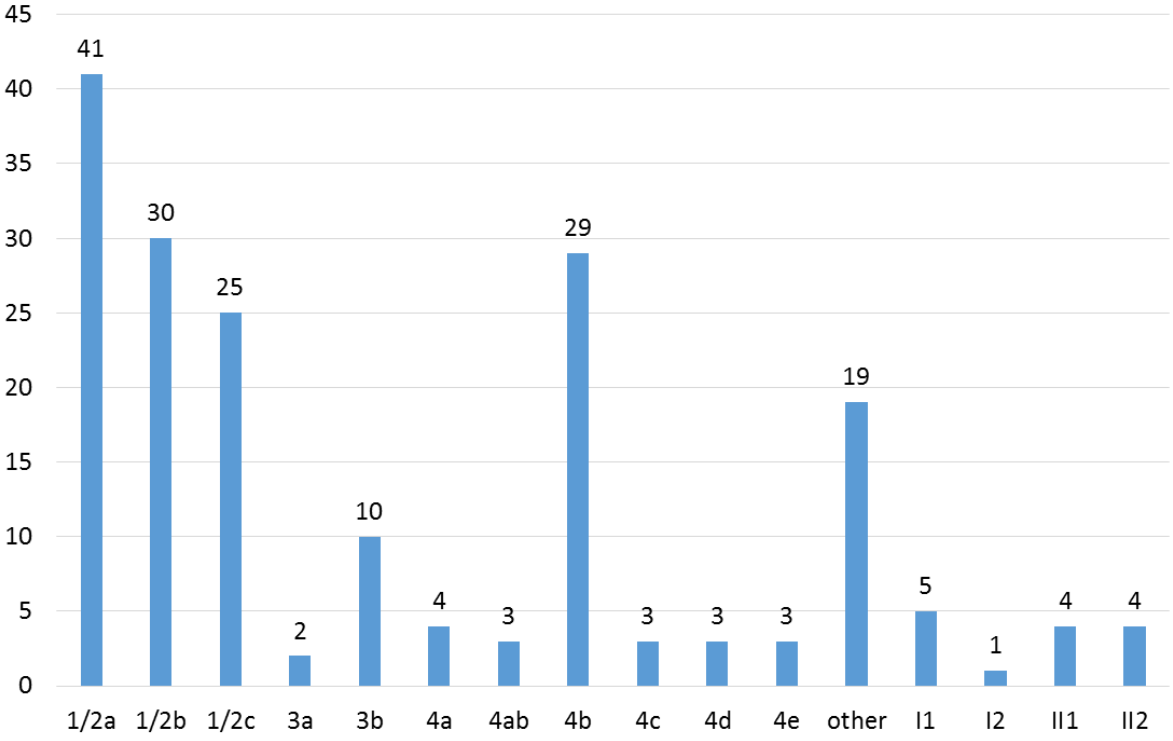
# III. Results and Outputs

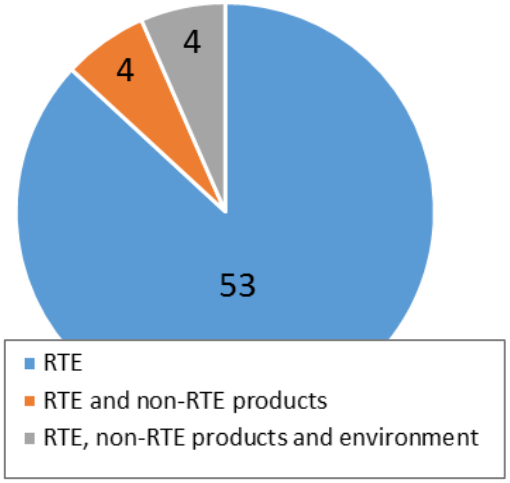
20% of studies provided serotype information

Results about the reported ***Listeria monocytogenes* serotypes or lineages**, number indicates the number of times that the serotype was detected and reported among the eligible records

- **Number of isolates** analysed by article: **1 to 1280**

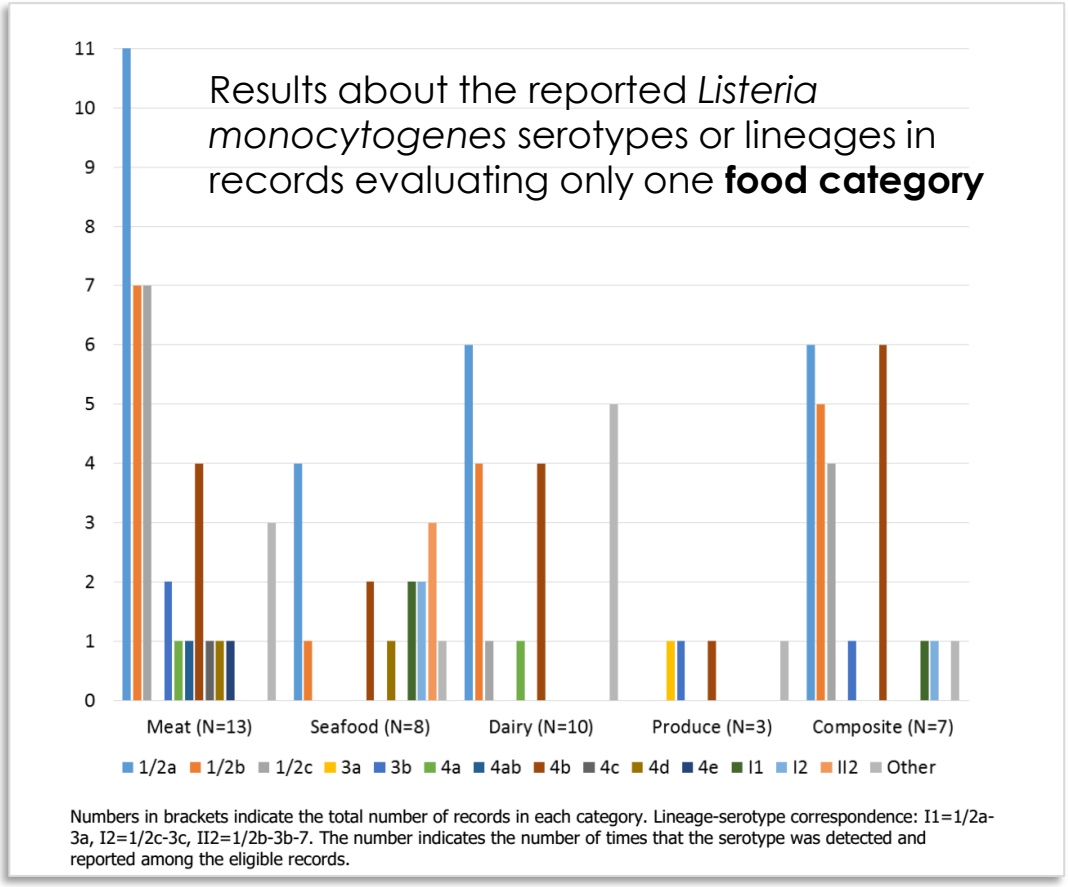
- **Origin of the isolates**




Lineage-serotype correspondence:
**I1**=1/2a-3a, **I2**=1/2c-3c, **II1**=4b-4d-4e and **II2**=1/2b-3b-7

# III. Results and Outputs

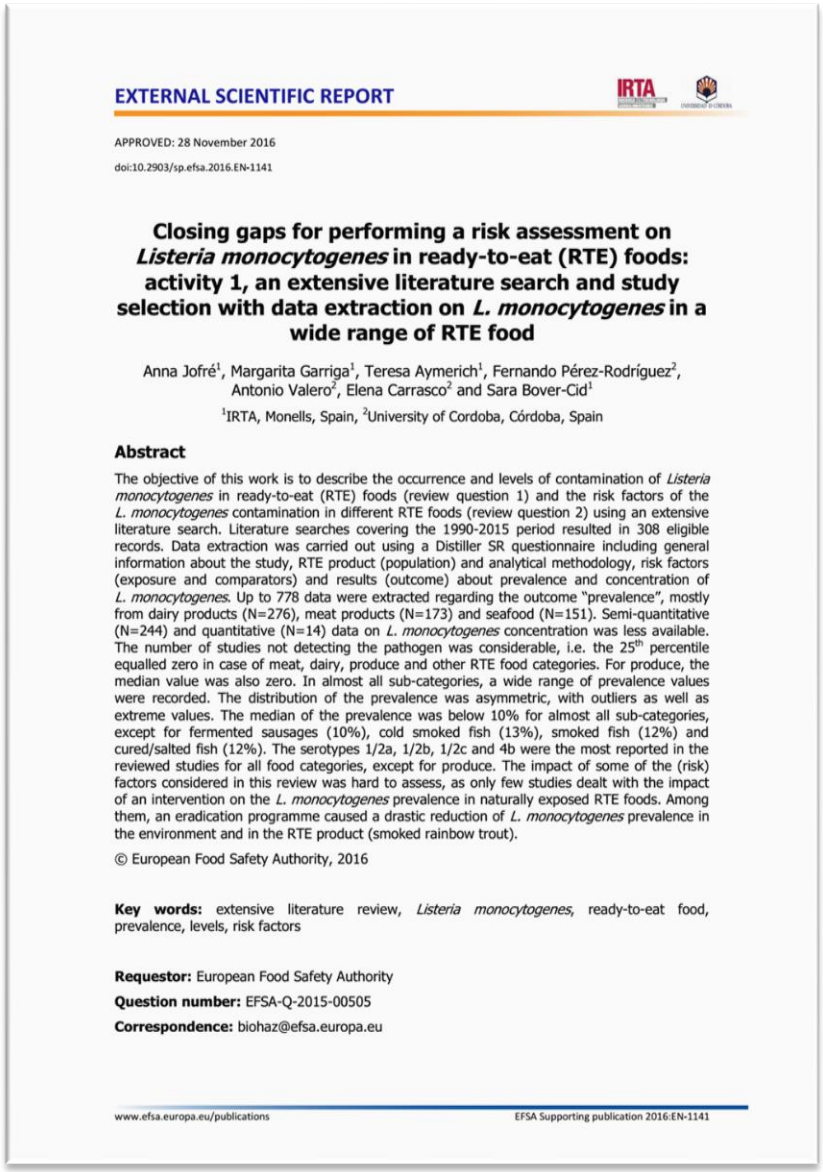



Numbers in brackets indicate the total number of records in each category. Lineage-serotype correspondence: I1=1/2a-3a, I2=1/2c-3c, II2=1/2b-3b-7. The number indicates the number of times that the serotype was detected and reported among the eligible records.

# III. Results and Outputs

**4** to **synthetize data** to reach the general objectives

Anna Jofré, Margarita Garriga, Teresa Aymerich, Fernando Pérez-Rodríguez, Antonio Valero, Elena Carrasco and Sara Bover-Cid, 2016. Closing gaps for performing a risk assessment on *Listeria monocytogenes* in ready-to-eat (RTE) foods: activity 1, an extensive literatura search and study selection with data extraction on *L. monocytogenes* in a wide range of RTE food. EFSA supporting publication 2016:EN-1141. 184 pp. doi:10.2903/sp.efsa.2016.EN-1141



EXTERNAL SCIENTIFIC REPORT

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**Closing gaps for performing a risk assessment on *Listeria monocytogenes* in ready-to-eat (RTE) foods: activity 1, an extensive literature search and study selection with data extraction on *L. monocytogenes* in a wide range of RTE food**

Anna Jofré[1], Margarita Garriga[1], Teresa Aymerich[1], Fernando Pérez-Rodríguez[2], Antonio Valero[2], Elena Carrasco[2] and Sara Bover-Cid[1]

[1]IRTA, Monells, Spain, [2]University of Cordoba, Córdoba, Spain

**Abstract**

The objective of this work is to describe the occurrence and levels of contamination of *Listeria monocytogenes* in ready-to-eat (RTE) foods (review question 1) and the risk factors of the *L. monocytogenes* contamination in different RTE foods (review question 2) using an extensive literature search. Literature searches covering the 1990-2015 period resulted in 308 eligible records. Data extraction was carried out using a Distiller SR questionnaire including general information about the study, RTE product (population) and analytical methodology, risk factors (exposure and comparators) and results (outcome) about prevalence and concentration of *L. monocytogenes*. Up to 778 data were extracted regarding the outcome "prevalence", mostly from dairy products (N=276), meat products (N=173) and seafood (N=151). Semi-quantitative (N=244) and quantitative (N=14) data on *L. monocytogenes* concentration was less available. The number of studies not detecting the pathogen was considerable, i.e. the 25th percentile equalled zero in case of meat, dairy, produce and other RTE food categories. For produce, the median value was also zero. In almost all sub-categories, a wide range of prevalence values were recorded. The distribution of the prevalence was asymmetric, with outliers as well as extreme values. The median of the prevalence was below 10% for almost all sub-categories, except for fermented sausages (10%), cold smoked fish (13%), smoked fish (12%) and cured/salted fish (12%). The serotypes 1/2a, 1/2b, 1/2c and 4b were the most reported in the reviewed studies for all food categories, except for produce. The impact of some of the (risk) factors considered in this review was hard to assess, as only few studies dealt with the impact of an intervention on the *L. monocytogenes* prevalence in naturally exposed RTE foods. Among them, an eradication programme caused a drastic reduction of *L. monocytogenes* prevalence in the environment and in the RTE product (smoked rainbow trout).



**Key words:** extensive literature review, *Listeria monocytogenes*, ready-to-eat food, prevalence, levels, risk factors

**Requestor:** European Food Safety Authority

**Question number:** EFSA-Q-2015-00505

**Correspondence:** biohaz@efsa.europa.eu

# IV. Conclusions

The study provides a detailed description of the extensive **literature searches** on the occurrence and levels of contamination of *Listeria monocytogenes* in RTE foods (time period 1990-2015).

The distribution of the reported **prevalence** is asymmetric, with several outliers as well as extreme values (being highest in seafood and cooked meat products). Prevalence equal to zero was reported in 29.5%, 21.9%, 41.2%, 63.5% and 52.9% of the studies dealing with meat products, seafood, dairy products, produce and other, respectively.

The impact of the **(risk) factors** considered in this review is hard to be assessed, as the studies usually do not provide the outcome (prevalence and/or level values) as a function of the risk factors.

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