anses
French agency for food, environmental and occupational health safety

Maisons-Alfort laboratory for food safety

EURL Lm
European Union Reference Laboratory for *Listeria monocytogenes*

2013 Work Programme of the European Union Reference Laboratory for *Listeria monocytogenes*

Version 3 – 19 November 2012

French Agency for Food, Environmental and Occupational Health Safety – Maisons-Alfort Laboratory for Food Safety
23, avenue du Général de Gaulle - F94706 Maisons-Alfort Cedex - Telephone: + 33 149 771 300 - Fax: + 33 143 689 762 - www.anses.fr

INTRODUCTION

In May 2006, the Maisons-Alfort laboratory for food safety of Anses (French agency for food, environmental and occupational health safety) has been nominated European Union Reference Laboratory for *Listeria monocytogenes* (EURL *Lm*) (see EC Regulation 776/2006).

The EURL *Lm* foresees to undertake the following actions in 2013, according to the actions planned at the 6th Workshop of the National Reference Laboratories (NRLs) (28-30 March 2012).

Most of these activities aim at implementing, from an analytical point of view, the EC Regulation 2073/2005 on microbiological criteria for foodstuffs, which includes in particular 4 food safety criteria on *L. monocytogenes* (Annex I, Chapter 1):

- either qualitative criteria: absence of *L. monocytogenes* in 25 g, for
  - ready-to-eat foods intended for infants and for special medical purposes,
  - other ready-to-eat foods able to support the growth of *L. monocytogenes*, when leaving the producer;
- either quantitative criteria: a limit of 100 cfu/g, for
  - ready-to-eat foods able to support the growth of *L. monocytogenes*, placed on the market during their shelf-life,
  - ready-to-eat foods unable to support the growth of *L. monocytogenes*, placed on the market during their shelf-life.

In addition, Article 5 (paragraph 2) of EC Regulation 2073/2005 requests that:

- Samples shall be taken from processing areas and equipment used in food production, when such sampling is necessary for ensuring that the criteria are met. In that sampling the ISO standard 18593 shall be used as a reference method;
- Food business operators manufacturing ready-to-eat foods, which may pose a *L. monocytogenes* risk for public health, shall sample the processing areas and equipment for *L. monocytogenes* as part of their sampling scheme.

NB 1: In brackets under each item, the scheduled duration of the action is indicated: either annual (limited to 2013), either multi-annual (on-going programme on several years).

NB2: The activities are gathered according to the tasks allocated to EURLs, defined EC Regulation 882/2004 on official controls (Article 32, paragraph 1 on EURLs for feed and food):

- *Section 1: Dispatch of methods and proficiency testing trials for the NRLs,*
- *Section 2: Analytical development,*
- *Section 3: Training of the NRLs,*
- *Section 4: Technical and scientific assistance to the European Commission.*

0 GENERAL ASPECTS

0.1 GENERAL COORDINATION (MUTI-ANNUAL)

General coordination by the EURL (management team, administrative department -PAFT) of the NRL network (dispatch of circular letters and documents, coordination of the scientific and technical support to NRLs,…).

In particular, the EURL deputy manager has undertaken a visit of the NRLs, in order to better know them, their teams, premises, and exchange on their NRL missions and activities, as well as on their expectations from the EURL.
1 mission in 2013: 1 visit of closely located NRLs.

Relations with DG SANCO, coordination of the scientific and technical advice to DG SANCO, management of annual contract with DG SANCO (annual budgets and work programmes, annual technical and financial reports).

In-house follow-up of EURL activities, expenses, support to laboratory units involved in EURL activities.

0.2 WORKSHOP OF THE NRLS (ANNUAL)

The EURL will organize the 7th Workshop of the NRLs in 2013, of general scope:

- to make a progress report on works undertaken by the EURL since the 2012 Workshop;
- to envisage the work programme for 2014 and later.

Three experts would be invited, as well as NRLs from accessing countries.

0.3 SCIENTIFIC MONITORING AND COMMUNICATION (MULTI-ANNUAL)

The EURL teams will review scientific publications in the EURL area of competence, as well as communicate on the works conducted as EURL Lm, disseminate the outcome of works in the international scientific community (drafting of written publications, oral presentations and posters to international symposia).

1 DISPATCH OF METHODS AND PROFICIENCY TESTING TRIALS

1.1 DETECTION AND ENUMERATION OF *L. MONOCYTOGENES* IN FOOD

1.1.1 STUDY OF SAMPLE TYPES FOR INTER-LABORATORY TRIALS (MULTI-ANNUAL)

Duration: started: 2011 – expected end: 2013

Objective

The EURL *Lm* (Unit EDB) is studying a variety of ready-to-eat (RTE) food matrices to be used as samples for proficiency testing (PT) trials on *Lm* detection and enumeration, as to be representative of the broad scope of the *Lm* microbiological criteria in all RTE foods. Indeed, NRL ability to implement the reference methods for *Lm* detection and enumeration (EN ISO 11290-1&2) may vary, depending on the type of food matrices analysed (sample preparation). Up to now, the EURL has investigated the following sample types: a liquid matrix (pasteurized milk), a powdered matrix (powdered infant formula), a preserved fish product (cold-smoked salmon) and a fresh meat product (diced poultry). Apart from the strategy to cover the scope of the microbiological criteria on *Lm*, this choice of matrices took account of requests from NRLs.

Expected output and time of delivery

The EURL *Lm* will complete in 2013 the study to develop the sample types to be used for PT trials, using as matrix diced poultry, with possibly the addition of a competitive microflora. In particular, the homogeneity and stability of this sample type will be studied.

Relation to EURL specific tasks (article 32.1 of EC Regulation 882/2004): b.

1.1.2 ENUMERATION OF *L. MONOCYTOGENES* (ANNUAL)

Objective

PT trials organised by the EURL *Lm* (Unit EDB) for the NRLs *Lm* aim at evaluating the ability of the NRLs to apply satisfactorily the reference method EN ISO 11290-2 for the enumeration of *L. monocytogenes*, in the frame of controls prescribed by EC Regulation 2073/2005.

Expected output and time of delivery

The EURL *Lm* will organize in 2013 a PT trial for the NRLs on the target described before, using diced poultry as matrix.

Relation to EURL specific tasks (article 32.1 of EC Regulation 882/2004): b.

1.1.3 COMPARISON OF INOCULATION TECHNIQUES OF SOLID FOOD MATRICES (MULTI-ANNUAL)

Duration: started: 2011 – expected end: 2014

Objective

Solid food matrices are part of RTE food included in the microbiological criteria for *Lm*, in EC Regulation 2073/2005, thus it is necessary to organize PT trials on solid food matrices. The EURL *Lm* (Unit EDB) has already organized PT trials on such sample types, but the artificial contamination technique should be optimized. In particular the inoculum should be more standardised in order to obtain more precise levels, which is essential at very low levels. The most appropriate competitive strains of *Listeria* species, in mixture with *Lm*, should be also investigated for use to contaminate samples for PT trials. In fact a risk of overgrowth phenomena may occur during the enrichment phase of the detection method.

In addition, the EURL has organized PT trials up to now without including sub-sampling of test portion: the possibility to include this initial step of the analysis (which is essential for a reliable analysis of solid matrices) will be tested.

Expected output and time of delivery

After having conducted in 2012 a bibliographic review, and launched an enquiry to the NRLs to collect their experience as PT trial organisers at national level, the EURL *Lm* (Unit EDB) will test and compare in 2013-2014 different inoculation techniques of solid food matrices, in-depth or in surface, so as to optimize the combination between the solid food matrix and the inoculation technique. In particular, the EURL will study the homogeneity and stability of these newly developed sample types.

If satisfactory results would be obtained, this study would be used by the EURL *Lm* for future PT trials and could help NRLs for the organization of their inter-laboratory PT trials at national level.

1 mission: visit of a technical center performing possible contamination techniques.

Relation to EURL specific tasks (article 32.1 of EC Regulation 882/2004): b.

1.2 SHELF-LIFE STUDIES RELATED TO *L. MONOCYTOGENES*

Frame: The EC Regulation 2073/2005 on microbiological criteria defines a quantitative limit for *L. monocytogenes* of 100 cfu/g, which is applicable to certain categories of products placed on the market during their shelf-life. The manufacturer needs to be able to demonstrate, to the satisfaction of the national Competent Authority (CA), that the product will not exceed the limit of 100 cfu/g throughout the shelf-life of the product. For that purpose, Annex II of the regulation lists the different types of data and studies that can be used.

1.2.1 REVISION OF TECHNICAL GUIDANCE DOCUMENT ON SHELF-LIFE STUDIES (MULTI-ANNUAL)

Duration: started: 2012 – expected end: 2013

Objective

In November 2008, the "EURL *Lm* Technical Guidance Document on shelf-life studies for *L. monocytogenes* in ready-to-eat foods" (2nd version), prepared by the EURL *Lm* in collaboration with a working group of 6 NRLs, was published and approved by EU Member States. It describes the microbiological procedures for determining growth of *L. monocytogenes* using challenge tests and durability studies, which are quoted in Annex II of EC Regulation No 2073/2005.

Expected output and time of delivery

The EURL *Lm* (Unit MOB) has initiated in 2012 a revision of the EURL *Lm* Technical Guidance Document and foresees to finalise it in 2013, with the collaboration of an extended NRL working group, which first met on 1-2 October 2012.

This revision takes into account:

(i) new scientific literature published on challenge-tests and durability studies,
(ii) experience of laboratories performing such studies and laboratory tests,
(iii) outcome of the EURL *Lm* study on growth variability of a collection of *L monocytogenes* strains,
(iv) proposals and feedback from NRLs, in recent years.

Meeting: 5 participants with only one with transportation costs (WG meeting combined with the 2013 annual workshop).

Relation to EURL specific tasks (article 32.1 of EC Regulation 882/2004): e

1.2.2 PROTOCOL FOR PT TRIALS ON CHALLENGE TESTS ASSESSING THE GROWTH POTENTIAL (MULTI-ANNUAL)

Duration: started: 2012 – expected end: 2013

Objective
The EURL *Lm* intends to organize PT trials for the NRLs, to evaluate their ability to apply satisfactorily the EURL *Lm* Technical Guidance Document on challenge tests assessing *Lm* growth potential. For that purpose, it is necessary to define at first a protocol for this type of PT trials: to our knowledge, PT trials for this type of studies have never been organized; no reference document for their organization is available.

Expected output and time of delivery
In 2013, the EURL *Lm* (Unit MOB), in collaboration with the same working group constituted to revise the EURL *Lm* Technical Guidance Document, will draft a protocol to organize PT trials on challenge tests assessing the growth potential.

Relation to EURL specific tasks (article 32.1 of EC Regulation 882/2004): b

1.2.3 TECHNICAL ASSISTANCE TO NRLS AND DISPATCH OF STRAINS (MULTI-ANNUAL)

Upon request of the NRLs, the EURL *Lm* (Unit MOB) would provide technical and scientific assistance, in particular on shelf-life studies for *Lm* in ready to eat foods.

Relation to EURL specific tasks (article 32.1 of EC Regulation 882/2004): a

The EURL *Lm* (Unit MOB) has completed in 2012 the development and characterization of a set of 24 strains to perform challenge tests in the frame of the use of the Technical Guidance Document on shelf life studies.

In 2013, the EURL will dispatch this set of strains upon request of the NRLs, whose transportation will be subcontracted.

Relation to EURL specific tasks (article 32.1 of EC Regulation 882/2004): e

1.3 LM STRAIN CHARACTERIZATION AND TYPING

1.3.1 CENTRAL EUROPEAN MOLECULAR DATABASE ON FOOD *L. MONOCYTOGENES* ISOLATES (MULTI-ANNUAL)

Frame: A European database on *L. monocytogenes* strains isolated from food would represent an efficient tool for detecting *Lm* clusters at European level and investigating the food origin of human listeriosis clusters/epidemics. This tool has been developed by the EURL *Lm*, in close collaboration with ECDC and EFSA.
DG SANCO has planned that a central database on molecular typing and associated epidemiological data, related to several pathogenic bacteria, including *Lm*, should be settled at EFSA, with a key role of the concerned EURLs as curators of the database. Once this database would be created, the EURL *Lm* database would be transferred to EFSA. In the meantime (2013), the EURL Lm would be used.

1.3.1.1 EURL *LM* DB

Objectives

The EURL *Lm* (Unit CEB), together with a Steering Committee (SCOM) composed of representatives from 8 NRLs, EFSA and ECDC, has developed since 2011 a European database on *L. monocytogenes* strains isolated from food (PFGE molecular sub-typing profiles and associated epidemiological data), named EURL *Lm* DB.

Expected output and time of delivery

Pilot submissions will be conducted in 2013.

In 2013, the EURL *Lm* DB will be used to centralise the PFGE profiles of the strain isolated during the European baseline survey: see 1.3.1.2.
Transport of strains from some NRLs to EURL will be subcontracted.

Database maintenance and update of Bionumerics software will be subcontracted.

The EURL *Lm* will strengthen its collaboration with PulseNet international network through its participation to the PulseNet international strategic plan, and will actively collaborate with PulseNet partners as well as continue its collaboration with ECDC, in case of *Lm* international alerts or for research collaboration.
Missions: 1 missions to US or Canada, 1 mission to ECDC.

The EURL will organize the 3rd SCOM meeting, together with the 2013 annual workshop.

1.3.1.2 EUROPEAN LM BASELINE SURVEY (BLS) IN RTE FOOD PRODUCTS: SUB-TYPING OF FOOD ISOLATES

Objective

Further to the European monitoring programme on the prevalence of *Listeria monocytogenes* in certain ready-to-eat foods ("baseline study" –BLS-, see EC Decision 2010/678 of 5 November 2010) and at the condition that EU Member States give their agreement, the EURL *Lm* will be involved, in collaboration with EFSA and ECDC, in a joint typing study of the strains isolated from the BLS and from patients during the same period.

Expected output and time of delivery

The EURL for *Lm* (Unit CEB) will coordinate the NRL network as a consortium of laboratories for the typing, conducted during the 2nd half of 2012/beginning of 2013. Two cases will be encountered:

- For the NRLs having successfully participated to the EURL PT trials on PFGE & molecular serotyping: they will type the strains isolated in their respective countries, and the EURL will interpret the data. For the NRLs which as SCOM members, they will submit their data online and the EURL will be curator of these data.
- For the NRLs which don't have the capacity to type the strains or whose proficiency to perform PFGE and/or molecular serotyping has not been assessed: they will send the BLS strains isolated in their respective countries to the EURL which will type them. The transportation of the strains from NRLs to the EURL will be sub-contracted.

Since the EFSA central database won't be settled at the time of this study, the typing data from the study will be entered in the EURL *Lm* database and will be transferred to the EFSA database, once created.

During the same period, the EURL will validate its PFGE protocol, in collaboration with the human health laboratory (SSI, DK, selected by ECDC) in order to confirm the full compatibility of the EURL and CDC protocols.

In 2013, the EURL will also play an active role in the technical management of the EDC/EFSA/ EURL *Lm* joint typing study. The EURL will be involved in the PFGE typing data interpretation and evaluation.

Relation to EURL specific tasks (article 32.1 of EC Regulation 882/2004): e

1.3.2 TECHNICAL & SCIENTIFIC ASSISTANCE TO NRLS, DISPATCH OF STRAINS (MULTI-ANNUAL)

Upon their request, the EURL *Lm* (Unit CEB) would provide technical and scientific assistance to NRLs (in particular to perform PFGE and PCR methods), and would send them *Lm* field strains from its collection, as well as the control strains *Salmonella* Braenderup H9812 and *L. monocytogene*s H2446.

The EURL *Lm* (Unit CEB) will update its in-house methods for PFGE, molecular serotyping and conventional serotyping, and dispatch them to the NRLs. The new version of these in-house methods will include the recent evolutions of the international protocols and the improvements made by the EURL during the past years.

Mission: 1 mission to the PL-NRL for on-site assistance to implement PFGE and PCR techniques.

2 ANALYTICAL DEVELOPMENT

2.1 DETECTION AND ENUMERATION OF *L. MONOCYTOGENES* IN FOOD

2.1.1 ENUMERATION METHOD USING A MEMBRANE FILTRATION METHOD (MULTI-ANNUAL)

Duration: started: 2008 – expected end: 2013

Objective

The Standard horizontal method EN ISO 11290-2 for enumeration of *L. monocytogenes* in food is characterized by a theoretical limit of enumeration of 10 - 100 cfu/g or ml. Meanwhile, it has been shown that the precision of this Standard method is quite poor at low levels. Even if the Standard has been amended with a more precise method (the LOA enumeration agar is now more specific to *L. monocytogenes*), the method still lacks of enough sensitivity to control precisely a limit at 100 cfu/g or ml or lower.

The EURL *Lm* (Unit EDB) has developed and validated a more sensitive enumeration method than the EN ISO 11290-2 one, including a concentration step based on membrane filtration followed by transfer of the filter to a selective medium, for the enumeration of *L monocytogenes* at low levels. Since 2008, the EURL has tested the applicability of this membrane filtration method to various food categories, using naturally contaminated samples.

Expected output and time of delivery

As agreed at the 2012 Workshop, the applicability of the method will be further studied in 2013, for some food matrices where there are not enough data, by lack of naturally contaminated samples obtained: vegetables, some delicatessen, prepared dishes and pastries, and some seafood products such as molluscs, shrimps . Sample transportation from some NRLs to EURL will be subcontracted.

The study will go on in collaboration with different NRLs (Czech Republic, Cyprus and Norway), willing to test the method, or which have developed other *L. monocytogenes* enumeration protocols at low levels, based on a concentration step of the suspension to be analysed. Newly developed methods will be tested in parallel, using a protocol harmonized with the collaborating laboratories. A meeting will take place together with the annual NRL workshop.

The EURL *Lm* will also update its bibliographic study on the enumeration techniques for *L. monocytogenes* at low levels: in particular the alternatives to the membrane filtration method, which could be used in cases where the latter would not be applicable.

Relation to EURL specific tasks (article 32.1 of EC Regulation 882/2004): c

2.1.2 REDUCTION OF THE SECOND ENRICHMENT STEP IN FRASER BROTH FROM 48 TO 24H

Duration: started: 2010 – expected end: 2013

Objective

Several NRLs have pointed out the length of the Standard reference method for the detection of *L. monocytogenes* in food, EN ISO 11290-1, based on conventional microbiology and requiring in particular for this bacterium two successive enrichment steps (3 days are needed to get a negative result and 4-7 days are required for a positive result). It makes the method not optimal for obtaining results shortly in the frame of own checks or official controls.

The 2nd enrichment step of the Standard method EN ISO 11290-1 is conducted in Fraser selective broth, incubated for 48 h at 37°C.

The purpose of this study is to investigate the possibility to reduce the duration of the second enrichment step from 48 h to 24 h, which would enable to shorten the total duration of the Standard detection method by 24h, representing a significant improvement in its practicability.

The CEN/ISO working group, in charge of the revision of EN ISO 11290-1, has recommended CEN/TC 275/WG 6 to follow the EURL *Lm* recommendation to reduce the duration of the 2nd enrichment step to 24 h: the draft to be submitted to the CEN/ISO parallel enquiry should include this change.

Expected output and time of delivery

The EURL *Lm* (Unit EDB) has already conducted a study on the enrichment phases of the Standard detection method, suggesting that a 24h-incubation in Fraser broth could be sufficient to reach the maximum population, instead of the current practice of 48h. In 2012, the EURL went on this study on naturally contaminated samples from various origins.

However, to substantiate the reduction of the 2nd enrichment step, and according to the analysis which will be performed until the end of 2012, it may be required to analyse in 2013 additional naturally contaminated samples from various origins, for some food categories lacking of sufficient naturally contaminated samples (for example vegetables, milk products...). Such samples would be asked to NRLs. The transportation of samples from NRLs to the EURL will be sub-contracted.

Relation to EURL specific tasks (article 32.1 of EC Regulation 882/2004): c

2.1.3 MEASUREMENT UNCERTAINTY

Duration: started: 2011 – expected end: 2014

Objective

To conduct analyses for own checks and official controls related to the quantitative criteria on *L. monocytogenes* in ready-to-eat food defined in EC Regulation 2073/2005 modified (criteria 1.2 & 1.3 in Annex I, Chapter 1), it is important to know and to control the measurement uncertainty (MU) associated to the analytical results. For example, the result found may comply with the limit settled in the microbiological criterion (here 100 cfu/g) whereas the true result (lying in the uncertainty range) may not comply: in that case, a wrong interpretation of the result may be taken if ignoring MU. A correct interpretation of analytical results, in terms of conformity with regulatory limits, thus requires the knowledge of MU associated to these results as well as the limitation of this uncertainty as far possible.

In the series of Standards EN ISO 6887-2 to 5 on the preparation of test samples for microbiological analyses, it is not specified how to sub-sample the test portion in the laboratory sample (sample that is sent to the laboratory), depending on the different types of food matrices to be submitted to microbiological analyses. This stage is however recognized as a major MU source, in particular for solid matrices characterized by heterogeneous bacterial contaminations, such as matured cheeses, smoked fishes or meat products.

The EURL *Lm* (Units EDB and MOB) has launched a study to assess the impact of test portion sub-sampling on MU, in order to evaluate heterogeneity of contamination, to harmonize how to sub-sample test portions and to reduce MU. The purpose of this study is to harmonize the procedure of sub-sampling the test sample in solid matrices, thus (i) reducing the overall MU, and (ii) better ensuring that the contamination of a sample is correctly reflected in the test portion taken and analyzed.

The outcome of this study would be transferred to ISO (i) to provide data for the revision of ISO/TS 19036 (MU estimation for quantitative determinations), to quantify the MU part liked to sub-sampling of test portions, for solid matrices, and (ii) to revise EN ISO 6887 series to better define the procedure of sub-sampling the test portion in solid matrices.

Expected output and time of delivery

After having studied contamination heterogeneity and having modeled it, the EURL will launch in 2013 an experimental study on sub-sampling, in order to validate the results obtained by modelling: various sample size portions will be studied. The types of matrices studied will depend on samples made available, characterized by high weight and high levels of natural contamination. Consequently any type of matrices with these characteristics will be used. These characteristics are expected to be encountered for seafood, meat and cheese matrices.This would allow selecting the more pertinent test portion size, as to obtain a satisfactory MU associated with the sub-sampling technique.

This study requires the analysis of naturally contaminated samples from various origins, and could be conducted in collaboration with some NRLs. A call for samples has been launched at the 2012 Workshop. The transportation of samples from NRLs to the EURL will be sub-contracted.

Relation to EURL specific tasks (article 32.1 of EC Regulation 882/2004): a & c

2.1.4 ALTERNATIVE TECHNIQUES FOR CONFIRMATION STAGE

Duration: started: 2013 – expected end: 2014

Objective

Several NRLs have underlined that the confirmation stage of the Standard methods for detection and enumeration of *L. monocytogenes* (EN ISO 11290-1&2) is laborious and long to implement in routine use, for large scale own checks or official controls. This confirmation is based on biochemical tests, haemolysis reaction and CAMP test (inoculation of test and reference cultures). The NRLs wished to have an overview, for confirmation purposes, on alternative methods, in particular PCR methods.

Expected output and time of delivery

Further to the 2011 and 2012 Workshops, the EURL *Lm* (Unit EDB) will undertake in 2013 a bibliographic review on alternatives to *Lm* confirmation tests included in EN ISO 11290, and will review in particular their validation status. This study is planned to be performed in collaboration with some NRLs, in particular the UK- NRL.

In addition, the EURL *Lm* would conduct an enquiry to NRLs on their practices regarding this topic.

Relation to EURL specific tasks (article 32.1 of EC Regulation 882/2004): a

3 TRAINING OF THE NRLS

Upon request, the EURL could receive NRLs for individual training on specific topics.

3.1 SHELF-LIFE STUDIES RELATED TO *L. MONOCYTOGENES*

At the condition that the revision of the EURL *Lm* Technical Guidance Document on shelf-life studies for *L. monocytogenes* in ready-to-eat foods would be completed (see 1.2.1) and the revised version be approved, the EURL *Lm* (Unit MOB) would organize for the NRLs, at the end of 2013, a training session dedicated to the implementation of challenge tests and durability studies of *L. monocytogenes* in food, according to the revised version of the Technical Guidance Document.

Relation to EURL specific tasks (article 32.1 of EC Regulation 882/2004): d

3.2 STRAIN CHARACTERISATION AND TYPING

The EURL *Lm* (Unit CEB) will organize in 2013 a training session dedicated to sub-typing by PFGE. This session will include technical and theoretical courses and will take place at EURL.

Relation to EURL specific tasks (article 32.1 of EC Regulation 882/2004): d

4 TECHNICAL AND SCIENTIFIC ASSISTANCE TO THE EUROPEAN COMMISSION

4.1 DG SANCO ACTIVITIES (MULTI-ANNUAL)

Upon request of the services of DG SANCO in charge of food hygiene:

- Participation of the EURL manager (Bertrand LOMBARD), for the analytical aspects, to the update of Regulation 2073/2005 on microbiological criteria related to *Lm* (in particular to the possible addition to the existing criteria on *Lm*) and to the corresponding meetings of the MS WG on microbiological criteria
  Missions: 2 meetings, Brussels;
- Technical and scientific assistance of the Unit MOB for the implementation of the Annex II on studies to verify compliance with the 100 cfu/g-ml limit at the end of shelf-life;

and any new question which may arise during the year.

4.2 PARTICIPATION TO CEN/ISO STANDARDISATION ACTIVITIES (MULTI-ANNUAL)

On behalf of the EURL *Lm* and as EC representative:

- Participation of the EURL *Lm* manager (Bertrand LOMBARD) to the activities of ISO/TC 34/SC 9[1] & CEN/TC 275/WG 6[2] in particular for aspects related to the standardization of reference methods for *L. monocytogenes;*
  Mission: 1 joint plenary meeting, Berlin, DE, June 2013.
- Leadership by an EURL *Lm* senior scientist (Nathalie GNANOU-BESSE) for the revision and validation by inter-laboratory studies of the EN ISO 11290-parts 1 & 2 Standard methods, in the frame of the CEN Mandate M/381. Leadership of the corresponding CEN/ISO working group
  (costs covered by the CEN Mandate, no cost for the EURL budget).

[1] Sub-Committee 9 « Microbiology » of Technical Committee 34 « Food products »

[2] Working Group 6 « Microbial Contaminants » of Technical Committee 275 « Food analysis – Horizontal methods »