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French agency for food, environmental and occupational health safety

Maisons-Alfort laboratory for food safety

EURL Lm
European Union Reference Laboratory for *Listeria monocytogenes*

2015 Work Programme of the European Union Reference Laboratory for *Listeria monocytogenes*

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INTRODUCTION

In May 2006, the Laboratory for Food Safety, Maisons-Alfort location, 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 2015, according to the actions planned at the 8th Workshop of the National Reference Laboratories (NRLs) (10-11 April 2014).

Scientific & technical activities of EURL *Lm* are mainly undertaken, in the laboratory, by:

- Unit *Salmonella*, *E. coli* and *Listeria* (SEL) on *Lm* detection/enumeration in food and food processing environment, *Lm* ecophysiology, *Lm* characterization and epidemiosurveillance;
- Team Modelisation and Quantitative Risk Assessment (MOD-AQR) for predictive microbiology on *Lm*, in particular challenge tests and durability studies.

Most of these activities aim at implementing, from an analytical point of view, the EC Regulation 2073/2005 modified 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 2015), either multi-annual (on-going programme on several years).

NB2: The activities are gathered according to the tasks allocated to EURLs, as defined by 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: NRL training and support to 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 - SAG) of the NRL network (dispatch of circular letters and documents, coordination of the scientific and technical support to 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.

Missions:
2 missions at DG SANCO (Brussels, 1 day each)

0.2 WORKSHOP OF THE NRLS (ANNUAL)

The EURL will organize the 9th EURL/NRLs workshop in 2015, of general scope:

- to make a progress report on works undertaken by the EURL since the 2014 workshop;
- to envisage the work programme for 2016 and later.

This workshop will take place at EURL (Maisons-Alfort, France).

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

1 DISPATCH OF METHODS AND PROFICIENCY TESTING TRIALS

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

1.1.1 ENUMERATION OF *L. MONOCYTOGENES* IN RTE ICEBERG (ANNUAL)

Duration: 2015

Objective

PT trials organised by the EURL *Lm* (Unit SEL) 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 2015 a PT trial for the NRLs on *Lm* enumeration, using as matrix ready-to-eat iceberg salad.

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

1.1.2 CRITERIA FOR OUTSOURCING PART OF PT TRIALS AND FOR THE SELECTION OF SUB-CONTRACTORS

Duration: 2013 – expected end: 2016

Objective

Part of PT trials organized by NRLs for the national networks of official laboratories (OLs) may be outsourced, except follow-up of individual lab performance and corrective actions. NRLs highlighted the need of guidance on how to select PT providers, including steps of PT trials that can be outsourced, frequency, details on method used by participants. A collaborative work with other EURLs in the area of biological risks is envisaged to develop an harmonized approach.

Expected output and time of delivery

In 2015, EURL *Lm* will set up WG of NRLs volunteering to collaborate with EURL to draft a technical guidance document on this topic.

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

1.2 *L. MONOCYTOGENES* STRAIN CHARACTERIZATION AND TYPING

1.2.1 PT TRIAL ON PFGE *L. MONOCYTOGENES* SUB-TYPING FOR THE NRLS

Duration: 2014-2015.

Objective

This inter-laboratory proficiency testing (PT) trial, organised by the EURL *Lm* (Unit SEL), aims at evaluating the ability of volunteering NRLs to perform satisfactorily sub-typing of *L. monocytogenes* strains by conventional serotyping, molecular serotyping, PFGE (Pulsed Field Gel Electrophoresis) and PFGE profile interpretation.

Expected output and time of delivery

A panel of 11 strains will be chosen in close collaboration with SSI, DK (upon contract for ECDC). The EURL *Lm* PT trial will be synchronised with the SSI PT trial for clinical national reference laboratories. EURL *Lm* will dispatch the strains for this PT trial in October 2014.

In 2015, EURL *Lm* will collect and analyse results from all the participants. It will ensure appropriate follow-up of non satisfactory results. It will also produce a preliminary report for each of the participants, then the final report.

1.2.2 EUROPEAN MOLECULAR DATABASES ON FOOD *L. MONOCYTOGENES* ISOLATES (MULTI-ANNUAL)

Duration: 2011- until transfer of EURL *Lm* database to EFSA

Objectives

EURL *Lm* (Unit SEL), together with a Steering Committee (SCOM) composed of representatives from 8 NRLs, EFSA and ECDC, has developed since 2011 a European database, called EURL *Lm* DB. It includes typing data of strains isolated from food, feed, animal and environment, as well as associated epidemiological data.

In the context of a new database being developed by EFSA, called Molecular Typing Data Collection system (MTDC) (see 4.2), EURL *Lm* prepares the NRL network to submit their profiles to the future EFSA DB. EURL provides a support to NRLs for their own DB organisation and for improving the quality of their PFGE profiles. As the profiles submitted to EURL *Lm* DB are mirrored on each NRL DB, it would be easy for the NRLs to re-submit to EFSA MTDC their profiles already validated through EURL Lm DB.

For example, EURL *Lm* can assist NRLs by providing a conversion tool. This tool will make it possible to translate the epidemiological description associated to strain profiles from the EURL *Lm* DB into the EFSA scheme (SSD2: Standard sample description 2).

Furthermore, the evolution of the DB system to databases such as the one of US CDC, which will use whole genome Multi Locus Sequence Typing (wgMLST) technique, has to be foreseen.

1.2.2.1 EURL *LM* DATABASE

Expected output and time of delivery

Pilot submission of *Lm* profiles from NRLs, which are SCOM members, has been achieved in 2013, mainly in the frame of the European baseline survey (see 1.3.2.2). EURL *Lm* DB is now open to NRL network and is in production phase. To be able to submit profiles to EURL *Lm* DB, NRLs need to have successfully participated to one EURL PT typing trial and to have agreed on the EURL *Lm* DB Memorandum of Understanding. The submission of profiles is a valuable opportunity for the NRLs: they gain training to organize their own DB and to perform retrospective validation of their historical data.

EURL *Lm* intends to maintain the EURL *Lm* DB operational until the EFSA database will be operational. The exact moment to stop the EURL *Lm* DB will be agreed within the joint EFSA-ECDC Steering Committee, to be settled by the end of 2014 (see 4.2).

EURL *Lm* will organize the 4th SCOM meeting, together with the 2015 annual workshop.

Meeting

1 meeting of EURL *Lm* DB SCOM, together with the annual workshop.

1.2.2.2 HARMONIZATION OF TYPING METHODS

Duration: 2014 - not yet defined

Objectives

In the context of set up by EFSA of the Molecular Typing Data Collection system on *Lm*, STEC and *Salmonella* (see 4.2), the curation of the PFGE profiles has to be performed by each EURL according to SOPs harmonized with ECDC for the three pathogenic bacteria. The harmonisation of SOPs includes the PFGE SOPs but also the SOPs of the new method typing method, under development and to be based on whole genome sequencing.

The harmonisation will be based on the data from SOP comparison between SSI and EURL *Lm*, produced in the frame of the European Listeria Typing Exercise (ELiTE) study, conducted by ECDC in collaboration with EFSA and EURL *Lm*.

Expected output and time of delivery

The first version of the three PFGE SOPs on PFGE testing, PFGE profiles' interpretation and PFGE profiles' curation, have been developed in 2014, in the frame of the contract between EURL *Lm* and EFSA. In 2015, EURL *Lm* intends to revise them, as to reach a better

harmonization (i) with the SOPs for the 2 other bacteria (STEC and *Salmonella)* and (ii) with the SOP for human strains, in the frame of ELITE project.

In 2015, EURL *Lm* will further strengthen its collaboration with PulseNet (PN) international network through its participation to the PulseNet international strategic plan.

To date, the whole genome MLST (wgMLST) method is being developed for *Lm* by US CDC in a 6 month project. The implementation of wgMLST should be discussed and evaluated between the 3 EURLs, EFSA and ECDC from 2015 and onwards.

Missions

1 mission to the PN USA headquarters (CDC, Atlanta), to allow EURL to strengthen its collaboration with the origin and leader of PulseNet (PN) international network, to have a better knowledge of tools used by CDC and to progress in harmonization with clinical sector.

1 mission to SSI (DK, upon contract for ECDC) to harmonize the next version of EFSA and ECDC database SOPs.

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

1.3.1 UPDATING OF THE GUIDANCE DOCUMENT TO EVALUATE THE COMPETENCE OF LABORATORIES IMPLEMENTING CHALLENGE TESTS (MULTI ANNUAL)

Duration: 2015 - 2016

Objective

A guidance document to evaluate the competence of laboratories implementing challenge tests on growth potential was developed by EURL *Lm* with a working group of volunteering NRLs, and proposed to DG SANCO in 2012 (Version 0-03/02/2012).

In 2014, EURL *Lm* (Team MOD-AQR) dispatched a questionnaire to the NRLs (05/03/14) to get information on the need and use of this document The outcome was presented and discussed at the 2014 workshop: the answers highlighted that this guide was needed but not enough used. It was clarified that the target users of this guide should be Competent Authorities (CAs) or NRLs, if mandated by their CAs. It was agreed that the need of such a guide should be confirmed by CAs.

Expected output and time of delivery

Depending on the outcome of the consultation of CAs on the need of this guide, EURL *Lm*, with a WG of volunteering NRLs, would launch the revision of this guide, in order to take into account the last version of the EURL *Lm* "Technical guidance document for conducting shelf-life studies on *Listeria monocytogenes* in ready-to-eat foods" (Version 3 -06/06/2014).

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

2 ANALYTICAL DEVELOPMENT

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

2.1.1 MEASUREMENT UNCERTAINTY: INFLUENCE OF THE SIZE OF TEST PORTION

Duration: 2011 – expected end: 2016

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* has conducted a study to assess the impact on MU of (i) the procedure to sub-sample test portion and of (ii) size of test portion, in order to evaluate heterogeneity of contamination, to harmonize how to sub-sample test portions and to reduce MU. The purpose of this study was 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.

One of outcome of this study confirmed that enlarging the size of test portion is a key factor to reduce MU. This outcome has been transferred to ISO/TC 34/SC 9/WG 8, in charge of revising the Standard series EN ISO 6887 on preparation of test samples, initial suspension and decimal dilutions. These results should be included in an informative of EN ISO 6887-1.

EURL also presented for discussion the results of this study at the 2014 annual workshop. MU was significantly reduced with a test portion size of 100g, compared to the current 10g, but this size of test portion is hardly applicable to routine analysis. 25 g was considered more applicable in routine analyses, in particular for official controls.

Expected output and time of delivery

As agreed at the 2014 workshop, EURL *Lm* (Unit SEL) will coordinate a study to assess the impact of test portion size on MU, in particular by comparing 10g or 25g test portions with naturally contaminated samples. The purpose would be to introduce recommendations on test portion size (i) in the frame of the revision of EN ISO 11290-2 Standard for the specific case of *Lm* enumeration (recommendation to CEN/TC 275/WG 6/TAG 17 *Listeria,* if enough data available before revision of EN ISO 11290-2 is finalized), and (ii) in the next revision of EN ISO 6887-1 (recommendation to ISO/TC 34/SC 9/WG 8).

EURL will launch a call for participation of NRLs to this study.

Given the heaviness of this project and the difficulty to collect naturally contaminated samples, with high contamination levels, this study needs to be conducted over 2 years, and the expected end has to be extended to 2016.

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

2.1.2 APPLICABILITY OF EN ISO 11290-1&2 STANDARD METHODS FOR *L. MONOCYTOGENES* DETECTION & ENUMERATION IN PRESENCE OF NEW *LISTERIA* SPECIES

Duration: 2014 – expected end: 2015

Objective

During the past four years, new species of the genus *Listeria* have been isolated from foods and other environmental niches worldwide: *L. marthii, L. rocourtiae, L. fleischmannii, L. weihenstephanensis, L. floridensis, L. aquatica, L. Cornellensis, L. riparia*, and *L. grandensis*). It is not known whether the Standard methods EN ISO 11290-1&2 under revision (in particular their confirmation stage) can correctly differentiate *Lm* from these new species. There is a risk of lack of specificity of these methods (false positives).

Since the Standard methods under revision will include all other *Listeria* species in addition to *L. monocytogenes*, it is necessary to check the Standard methods' ability to recover and detect the newly identified *Listeria* species. In particular, certain characteristics of these newly discovered species remain unknown, such as: their growth and colony characteristics on commonly used *Listeria* selective isolation agars, such as LOA agar prescribed in EN ISO 11290-1&2, their reaction to some biochemical tests used for confirmation in the Standard methods, their growth performance in the selective enrichment broth of EN ISO 11290-1 in the presence or absence of other *Listeria* spp.

Expected output and time of delivery

EURL *Lm* (Unit SEL) will go on investigating the above-mentioned questions and test the ability of the current Standard methods to detect & enumerate *Lm* in the presence of these new species. Potential collaborations have to be confirmed.

The results of this study will be transferred to the CEN/TC 275/WG 6/TAG 17 *Listeria* for the revision of the Standard methods EN ISO 11290-1&2. In particular, a 3rd meeting of TAG 17 is planned in 2015, after the closure of the CEN/ISO parallel enquiry, in order to prepare the drafts to be submitted to CEN/ISO Final Votes. It would be important to have data available for this meeting. In fact, at this stage, and since the inter-laboratory studies organized in 2013 to validate the Standard methods concerned only *L. monocytogenes*, some modifications are still possible regarding detection or enumeration of *Listeria* spp. The study may also result in a more precise scope if the method would be unable to detect/enumerate some of the new species.
Relation to EURL specific tasks (article 32.1 of EC Regulation 882/2004): a, c

2.1.3 SAMPLE POOLING/COMPOSITING

Duration: 2015 – expected end: 2015

Objective

EN ISO 6887-1 Standard under revision on the preparation of test samples for microbiological analyses, will now include a general approach and experimental design to sample pooling/compositing (clause 9.3 and annex A), allowing to pool test portions or enrichment broths before subsequent analysis, as to reduce analytical costs, provided that an experimental study verifies that this practice has no impact on detection performance of the method. This study is quite heavy to perform for laboratories, since this study has to be conducted for each couple target/matrix, and a realistic stress has to be applied to the bacteria. Moreover, the study design is adequate to verify that pooling has no significant impact, or to identify a situation with heavy impact of pooling, but can hardly detect moderate effect on method performance.

At the 2014 workshop, NRLs wished that EURL conducts a study on impact of pooling to detect Lm according to EN ISO 11290-1.

Expected output and time of delivery

EURL *Lm* (Unit SEL) will investigate the impact of pooling samples on the performance of the Standard method EN ISO 11290-1, in collaboration with the National Veterinary School of Maisons-Alfort (ENVA). In particular, a modelisation of *L. monocytogenes* growth in the pre-enrichment broth of the detection method (half-Fraser) will be developed and validated, on the basis of previous studies conducted at the laboratory or ENVA with artificial or natural contamination. The model will then allow simulating different situations of pooling, and their impact on the percentage of positive samples detected. Simulations will be performed using realistic initial contamination levels, if possible distribution of prevalence and contamination levels found in the European baseline survey.

The results of this study will be transferred to CEN/TC 275/WG 6/TAG 17 *Listeria* for the revision of the Standard method EN ISO 11290-1, and may result in the addition of a note in the standard.
Relation to EURL specific tasks (article 32.1 of EC Regulation 882/2004): a, c

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

Frame: 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.

2.2.1 CHARACTERIZATION OF TEMPERATURE DISTRIBUTION IN EUROPE AT RETAIL LEVEL (ANNUAL)

Duration: 2015

Objective

According to the agreement within the EURL/NRL working group which prepared the last version of the EURL Lm Technical guidance document for conducting shelf-life studies on *Listeria monocytogenes* in ready-to-eat foods, EURL *Lm* (Team MOD-AQR) will take into account data from European baseline survey on *L. monocytogenes* to (i) get information on temperatures at retail in European countries, (ii) to simulate cold chain when assessing shelf-life of food, in any European country.

This study would allow in particular refining the temperature profile defined in the above mentioned EURL *Lm* guide to conduct a challenge test.

SCoFCAH has given its approval at its meeting of 16/06/2014 that EFSA transfers to EURL *Lm* these data.

Expected output and time of delivery

EURL *Lm* will conduct in 2015 this study, which will focus on:

- analysing temperature data for the 3 categories of food (smoked and gravid fish, soft and semi-soft cheese, packaged heat treated meat product) and for the different types of retailers (supermarkets, small shops, speciality delis, street markets) which were included in the European baseline survey;
- characterizing the temperature distribution at European level for each food and each retailer.

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

2.3 *L. MONOCYTOGENES* STRAIN CHARACTERIZATION AND TYPING

2.3.1 MOLECULAR SEROTYPING OF *L. MONOCYTOGENES*

Duration: 2014-2015

Objective

EURL *Lm* (Unit SEL) has already developed and dispatched in 2009 to the NRLs a protocol for molecular serotyping (by PCR) of *Lm*. In 2014, the EURL updated this protocol. However, new species of *Listeria* have been very recently described (see 2.1.2).

Expected output and time of delivery

EURL *Lm* will perform additional tests on the new *Listeria* species to ensure that the EURL *Lm* molecular serotyping scheme is specific of the species *L. monocytogenes*. If needed, EURL *Lm* will update its molecular serotyping protocol.

Subcontracting

Sequence verification will be needed and will require subcontracting for sequencing.

2.3.2 INVESTIGATION OF WHOLE GENOME SEQUENCING FOR TYPING OF *LISTERIA MONOCYTOGENES*

Duration: 2015 – expected end: not yet defined

Objective

Bacterial sequencing by Next Generation Sequencing (NGS)) is expected to become the best choice method in epidemiological typing. In order to fully explore the advantages and disadvantages of WGS-based data for *Lm* surveillance, EURL *Lm* and NRLs intend to set up a collaborative project. This project aims at developing harmonized SOPs for sampling, sample transportation, sample preparation, sequencing and collection of metadata. The strain selection would reflect different EURL/NRLs objectives, i.e. comparison of global diversity, detection of clones or clusters, detection of specific traits such as virulence or ecophysiology. This study should make it possible (i) to assess cutting-edge technology such as WGS and (ii) to compare it to the typing methods generally considered to be the standard methods at the national or international levels (). The whole project should enhance EURL/NRL network consortium expertise in methodology, as well as keep skills and analytical know-how abreast of those of its European and third-country partners.

Expected output and time of delivery

To conduct the above mentioned study, EURL *Lm* (Unit SEL) intends to generate WGS data on a collection of reference strains. This collection will include isolates well characterized by serotyping and PFGE, consolidated with extensive epidemiological meta-data associated to

these strains. The number of strains will be defined. This reference repository will be used for WGS validation and proficiency testing schemes for the NRL network.

In 2015, EURL intends to generate WGS data and to exchange these data between the partners, including volunteering NRLs.

Subcontracting

Shipment of strains from EURL to NRLs, or pick up of strains from NRLs to EURL.

Mission

One mission to the Italian NRL (IZSAM, Teramo) and one mission to the German NRL (BfR, Berlin) will be required.

Meeting

The EURL Lm will organize a first meeting with the volunteering NRLs This meeting may be scheduled in conjunction with the 2015 annual workshop.

3 TRAINING AND SUPPORT TO THE NRLS

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

3.1 STRAIN CHARACTERISATION AND TYPING

3.1.1 TRAINING SESSION (ANNUAL)

Objective, expected output and time of delivery

EURL *Lm* (Unit SEL) will organize in 2015 a training session dedicated to *Lm* sub-typing by PFGE. This session will include technical and theoretical courses and will take place at EURL.

EURL Lm intends to provide 2 on-site training sessions for 2 NRLs, which would require support in their PFGE database management.

Equipment

On-site training sessions will require running BioNumerics at the NRL. An internet license of BioNumerics is required to perform efficiently the training sessions.

Mission

Two missions for on-site training sessions in 2 NRLs.

3.1.2 TECHNICAL & SCIENTIFIC ASSISTANCE TO NRLS, DISPATCH OF SAMPLE AND/OR STRAINS (MULTI-ANNUAL)

Objective, expected output and time of delivery

a) EURL *Lm* (Unit SEL) will provide in 2015, in the frame of the EURL *Lm* DB project (see 1.3.2), technical remote assistance to the NRLs for:
   i) implementation of the database tools provided;
   ii) processing of their data at national level;
   iii) structuring of their epidemiological data.

   In the frame of the curation of the EURL *Lm* DB undertaken by EURL, the curator will ensure to the users a technical assistance by phone to solve the PFGE deviations observed in the NRL's profiles.

b) Upon NRL request, EURL *Lm* (Unit SEL) will provide technical and scientific assistance to NRLs, in particular to implement 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.

Sub contracting:

Dispatch of strains from EURL to NRLs

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

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

3.2.1 NRL TRAINING (ANNUAL)

Duration: 2015

Objective, expected output and time of delivery

EURL *Lm* (Team MOD-AQR, in collaboration with Unit SEL) will propose to the NRLs one training session (2 days) at EURL, taking into account the new version of the "Technical Guidance Document for conducting shelf-life studies on *L. monocytogenes* in ready-to-eat foods" (Version 3 -06/06/2014).

This session will include theoretical presentations, case studies and practical training.

This training session will focus on:

- challenge tests;
- durability studies;
- determination of shelf-life related to *L. monocytogenes*.

At maximum, 6 persons will be trained. If more applications would be received, a 2nd training session would be organized in 2016.

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 EC Regulation 2073/2005 on microbiological criteria related to *Lm* and to the corresponding meetings of the MS WG on microbiological criteria.

Missions:
3 meetings (1 day each, EC, Brussels).

4.2 TECHNICAL SUPPORT TO EFSA – CURATION OF EFSA DATABASE FOR LISTERIA MONOCYTOGENES

Duration: 2015- not yet defined

Objective

As requested by DG SANCO, EFSA is developing a central database (DB) on molecular typing for *Lm*, *Salmonella* and *E. Coli* (see DG SANCO vision paper[1]). This database, known as the EFSA Molecular Typing Data Collection system (MTDC), is prepared by EFSA since 2013, in collaboration with the concerned 3 EURLs. Each EURL will be the curator of the DB for the bacteria it is in charge of.

Expected output and time of delivery

In 2013-2014, EURL *Lm* has participated to the EFSA working group to develop MTDC and the associated DB. EFSA has also contracted with each of the 3 EURLs to develop and harmonize different SOPs.

The EFSA MTDC pilot will start in 2015. As planned by DG-SANCO and EFSA, each EURL will be curator of the PFGE profiles of its bacterium and will be in charge of the management of the scientific data. As planned in the EFSA MTDC draft technical report, the curation of the PFGE profiles will be performed in a central DB shared with ECDC, gathering clinical and non-clinical strain profiles.

The curation has to be performed in the same way between the 3 EURLs, for non-human strain profiles, and ECDC, for human strain profiles. This will require curation training sessions for these partners to harmonize the curation between them. EURL *E. coli* and *Lm* already propose a collaborative cross-validation between the curation teams of the 2 EURLs, needing the organisation of a curation "PT trial".

[1] "Vision paper on the development of data bases for molecular testing of foodborne pathogens in view of outbreak preparedness", approved by SCoFCAH on 12 December 2012.

EURL Lm will participate to the joint EFSA-ECDC Steering Committee, that will be in charge to manage the joint EFSA-ECDC DB, and which should be settled by the end of 2014.

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

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

- Participation of the EURL Lm manager (Bertrand LOMBARD) to the activities of ISO/TC 34/SC 9 [2] & CEN/TC 275/WG 6 [3] in particular for aspects related to the standardization of reference methods for L. monocytogenes;
  Mission: 1 joint plenary meeting, Netherlands, 22-26 June 2015.
- Leadership by a 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. Convenorship of the corresponding CEN/TC 275/WG 6/TAG 17 group on *Listeria*.
  (costs covered by the CEN Mandate, no cost for the EURL budget)
- Convenorship by two EURL *Lm* senior scientists (Brigitte CARPENTIER & Léna BARRE) of the new WG 17 of ISO/TC 34/SC 9, in charge of the revision of ISO 18593 on sampling techniques from food processing surfaces.
- Participation of a senior EURL *Lm* scientist (Annie BEAUFORT) to the new WG 19 of ISO/TC 34/SC 9, in charge of developing an EN ISO Standard on challenge tests in food and feed.
  Mission: 2 missions, 2 days each, in Europe.

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

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