



EUROPEAN HEALTH AND DIGITAL EXECUTIVE
AGENCY (HADEA)
Department A Health and Food Unit A2 EU4Health/SMP



Single Market Programme (SMP Food)

SMP-FOOD-2023-EURL-EURC-AG-IBA
Activities of the EU reference laboratories and EU
reference centres in 2023-2024

SUBMISSION FORM: DESCRIPTION OF THE ACTION
(Annex 1 – Description of the action (part B))

SMP-FOOD-2023-EURL-EURC-AG-IBA

Activities of the EU reference laboratories and EU reference centres in 2023-2024

Applicant shall provide information on each question contained in the Form. The information filled in the Form, shall be clear, concise, consistent and complete.

For questions on the information requested in this Form, please contact: please contact: HADEA-EURL@ec.europa.eu.

For questions on the [eGRANTS](#) Portal Submission System, please contact the [IT Helpdesk](#).

Applicant - COORDINATOR (Name of EURL)	Swedish Food Agency EURL for Foodborne Viruses
Topic	EURL for Foodborne Viruses SMP-FOOD-2023-EURL-EURC-AG-IBA
Implementation period	1/1/2023 – 31/12/2024

CONTACT PERSON for the programme :

Name	Magnus Simonsson
Function	EURL Director
e-mail	magnussimonsson@slv.se

ASSOCIATED PARTNER 1 (Name)	
Contact person :	
e-mail :	

Contents

1. LIST OF ABBREVIATIONS AND KEY WORDS.....	4
2. INTRODUCTION.....	4
3. ACTIVITIES.....	5
3.1. Work Package 1 - METHODS.....	5
Objectives: TO ENSURE AVAILABILITY AND USE OF HIGH QUALITY METHODS AND TO ENSURE HIGH QUALITY PERFORMANCE BY NRLs	5
3.2. Work Package 2 – ASSISTANCE TO NRLs.....	11
Objectives: TO PROVIDE SCIENTIFIC AND TECHNICAL ASSISTANCE TO NRLs	11
3.3. Work Package 3 – ASSISTANCE TO THE EUROPEAN COMMISSION AND OTHER ORGANISATIONS 15	
Objectives: TO PROVIDE SCIENTIFIC AND TECHNICAL ASSISTANCE TO THE EUROPEAN COMMISSION AND OTHER ORGANISATIONS.....	15
3.4. Work Package 4 - REAGENTS AND REFERENCE COLLECTIONS	20
Objectives:	20
3.5. REQUIREMENTS RELATED TO OTHER LEGISLATION	21
4. REMARKS.....	21

1. LIST OF ABBREVIATIONS AND KEY WORDS

HAV	Hepatitis A Virus
HEV	Hepatitis E Virus
LBM	Live Bivalve Molluscs
MS	Member State
NGS	Next Generation Sequencing
PT	Proficiency Testing
RT-dPCR	RT digital PCR
SOP	Standard Operating Procedure
SFA	Swedish Food Agency
WP	Work Programme

2. INTRODUCTION

The EURL has completed its fifth year and an organisation at the Swedish Food Agency (SFA) has continuously been built up. Moreover, a network of 30 National Reference Laboratories (NRLs) has been designated.

The network of NRLs has been established and in a complex area like foodborne viruses, several countries have designated two and even three laboratories.

Member states (MS) that still have not designated NRLs for foodborne viruses are: Croatia, Estonia (in the process to designate another MS), Greece (for non-animal matrices), Malta, and Portugal (in the process to designate). The Commission should encourage those countries to finalize their designation processes.

During 2020, EU decided to close the temporary legislation EU 2017/2298, amending EU669/2009, which stated increased level of official control of norovirus and hepatitis A virus (HAV) in frozen raspberries from Serbia. There are currently no microbiological criteria for foodborne viruses in the EU legislation, but a discussion about microbiological criteria for norovirus (and HAV) in bivalve molluscan shellfish, eaten raw, has been initiated by the Commission.

Annual PT distributions of molluscan shellfish spiked or bioaccumulated with norovirus and HAV are planned. Virus spiked soft fruits PT is also planned for the next coming years. Other matrices included in ISO 15216 have to be developed at the EURL. Swabbing techniques have been implemented and verification of swabbing of food surfaces are included in this WP. Further should bottled water be part of future PT schemes and implementation and verification of virus detection in bottled water is included in this WP. PT distribution of spiked bottled water is planned to 2024.

The standardisation process for hepatitis E virus detection and quantification in animal products follow its original plan and will probably be finalised during the duration of this WP.

The covid-19 pandemic has influenced the activities in the previous Work Programme. More or less all activities announced in the previous WP has been completed, sometimes postponed sometimes altered to a digital format instead of physical meetings. We hope that the pandemic not will influence the upcoming WP.

Regulation (EU) 625/2017 Art 94(2):

European Union reference laboratories designated in accordance with Article 93(1) shall be responsible for the following tasks insofar as they are included in the reference laboratories' annual or multiannual work programmes that have been established in conformity with the objectives and priorities of the relevant work programmes adopted by the Commission in accordance with Article 16 of Regulation (EU) No 2021/690:

(taking into account Art 147 of (EU) 625/2017)

3. ACTIVITIES

3.1. Work Package 1 - METHODS

Objectives: TO ENSURE AVAILABILITY AND USE OF HIGH QUALITY METHODS AND TO ENSURE HIGH QUALITY PERFORMANCE BY NRLs

- *Art. 94.2.a Providing national reference laboratories with details and guidance on the methods of laboratory analysis, testing or diagnosis, including reference methods.*
- *Art. 94.2.b Providing reference materials to national reference laboratories*
- *Art. 94.2.c Coordinating the application by the national reference laboratories and, if necessary, by other official laboratories of the methods referred to in point (a), in particular, by organising regular inter-laboratory comparative testing or proficiency tests and by ensuring appropriate follow-up of such comparative testing or proficiency tests in accordance, where available, with internationally accepted protocols, and informing the Commission and the Member States of the results and follow-up to the inter-laboratory comparative testing or proficiency tests.*

- *Art. 94.2.l Where relevant for their area of competence, cooperate among themselves and with the Commission, as appropriate, to develop methods of analysis, testing or diagnosis of high standards.*

Sub-activity 3.1.1 *Development, distribution of Proficiency tests (PT)*

Objectives:

To ensure harmonised analysis results among NRLs through the distribution of homogenous samples for PT and assess and report the results.

Description:

The ongoing discussions about microbiological criteria for noroviruses in oysters implies the importance to include this matrix in the PT scheme every year. Annual PT distributions are planned with bivalve molluscan shellfish matrix (2023 and 2024). In the previous years, there have been PT distributions of virus spiked oyster hepatopancreas (artificially spiked with HAV and norovirus, similar to what was done during validation of ISO 15216). We have started collaboration with a Swedish Marine Institute at the University of Gothenburg. Some permissions for the activity has not been approved, but we now seem to have solutions that will enable us to finalise the work. The Marine institute will be on a subcontract for setting up the facilities and provide localities. A study visit at the French NRL has served some education in the area and the EURL now implement alternative procedures and test bioaccumulation is planned for autumn 2023. Bioaccumulation in 2023 and 2024 means that there is a need for three persons to visit the facilities at two occasions yearly.

Questions were raised by NRLs regarding what method the EURL recommends for testing bottled waters and if it could be an option for PTs. As requested, bottled water will be distributed in 2024. Prior to the PT, development and verification of the method for bottled water will be carried out at the EURL (see 1.5).

To cover other matrix groups included in ISO 15216, soft fruits is planned to be distributed autumn 2023. As a request by the NRLs, this matrix group should be included in PT distributions at least every second year. The planned upcoming PTs are: soft fruits and oysters for 2023 and bottled waters and oysters in 2024.

Regarding the preparedness of staff and scientific and technical assistance to NRLs, the EURL will work towards a more quality assured and documented work flow e.g. for PT and to also eventually become accredited for PT according to ISO 17043.

For each new matrix an SOP will be produced, meaning that during the beginning of 2024 a new SOP describing techniques for virus extraction from bottled water will be produced and published on the EURL website.

The EURL has performed comparisons between RT digital PCR (RT-dPCR) and RT-qPCR in oyster matrix. The work has been published and RT-dPCR was concluded to be fit a possible alternative, especially if high precision is desired. During the workshop in 2018, the NRLs requested the inclusion of extra samples to assess variations of standard methods, such as the use of inhibitor removal kits and the use of RT digital PCR for the quantification of viral genomes.

A long-term goal is that RT-dPCR could be used as an alternative to the standard RT-qPCR described in ISO 15216. This should lead to more robust analyses and less variable results between laboratories.

However, for that, the method should be validated towards current standard method. An intra-laboratory study has been carried out during the last WP in collaboration with NRL Netherlands and eight other NRLs. A scientific publication is foreseen in 2023. Future PT distributions will give the possibility for the NRLs to provide results from both RT-qPCR and RT-dPCR. Continuous evaluation will add knowledge for the possibility to use RT-dPCR as an alternative to RT-qPCR.

Methods providing precise and accurate quantification of noroviruses in oysters are highly valuable for the implementation of future microbiological criteria.

Quality assurance of the EURL is important to maintain high quality and accreditation. The participation in external PT distribution is therefore of high importance. FAO Reference Centre for Bivalve Mollusc Sanitation, CEFAS, UK distribute PTs with norovirus and HAV contaminated Live Bivalve Molluscs (LBM). BIPEA is a European non-profit organization located in France. Gathering + 2600 member laboratories in the world (throughout 130 countries), it offers more than 220 regular proficiency testing programs. The EURL plans to participate in relevant parts of the proficiency testing schemes provided by these organisations. Costs are included in the budget.

Expected Output:

1. Available facilities and test for bioaccumulation of bivalves autumn 2023.
2. PT distribution of virus-contaminated raspberries, autumn 2023.
3. PT distribution of bioaccumulated oysters, november 2023.
4. Developed and verification of method for bottled water, spring 2024.
5. PT distribution of virus-contaminated bottled water, May 2024.
6. PT distribution of virus-bioaccumulated oysters November 2024.
7. Assessment and reports for each PT distribution and follow up of NRLs if necessary, 2023, 2024.
8. Quality assurance and maintained accreditation for the EURL through PT participation.

Duration:

2023 and 2024

Sub-activity 3.1.2 Implementation and development of typing methods for norovirus

Objectives:

To improve methods used for norovirus typing.

Description:

Typing methods of norovirus are of high importance both for tracing sources in outbreaks and to be able to follow the epidemiological development. The typing methods for norovirus are to some extent harmonised for clinical samples with classic Sanger sequencing, however for complex samples like food the methods cannot meet the requirements for inhibition and sensitivity.

Many noroviruses are so called recombinant viruses (one virus is a combination of two different types, polymerase type and capsid type). Norovirus type GII.4 Sydney 2012 is suggested to have had a

prolonged time as a pandemic virus due to such recombinations, thereby overcoming herd immunity. Many food samples, such as oysters, tend to harbour multiple types of norovirus. Conventional RT-PCR in combination with Sanger sequencing will only capture the most common type.

Next Generation Sequencing (NGS) makes it possible to find more or less all types of norovirus in a specific sample. With the current NGS technology, amplicon based sequencing has proven to facilitate the detection of more than one norovirus type in a single food sample. This will improve our capability for source attribution and tracing in outbreak situations, which, in turn, will help us to better understand the epidemiology, evolution and transmission of foodborne viruses. To type recombinant viruses, further development will be needed to sequence both the polymerase and capsid region. Promising testing has been done with NGS probe based capture chemistry and should be investigated further.

Typing through the cap region is implemented at the EURL and environmental samples has been analysed by NGS, showing the appearance of several genotypes in a single sample. In contrast Sanger sequencing will only be able to identify one single type. We will continue the work with the standard cap-sequence and a typing method for the cap region will be verified and harmonized, within the NRL network. Development of a sensitive pol-cap sequencing method using either probe-based capture chemistry or long read NGS to enable the identification of recombinant noroviruses will also be performed during this WP.

The use of probe-based capture chemistry also enables sequencing of full length genomes of noroviruses from complex samples and will be investigated by gathering experiences from other collaborating partners in the "EURL WG on NGS" and the "NRL WG on NGS".

There is also a need for general extended knowledge in both bioinformatics and platform competence to support NRLs in the start-up and use of NGS typing. We will continue the work to increase the EURL skills by collaboration with the "NRL WG for NGS" and the inter "EURL WG for NGS".

Expected Output:

1. A workflow for NGS targeting the cap region will be shared with the NRL WG on NGS, 2023
2. A verified and harmonized cap typing method on NGS, 2024
3. NGS technology has been developed for typing of multiple noroviruses in a sample, targeting the pol-cap-region. Enabling identification of recombinant noroviruses, 2024
4. Probe-based capture chemistry for whole genome sequencing of norovirus is investigated and experiences collected, 2023
5. Typing methods of noroviruses will be shared with NRLs.

Duration:

2023 and 2024

Sub-activity 3.1.3 Implementation and verification of method for detection and quantification on food surfaces according to ISO 15216.

Objectives:

To verify the ISO 15216 swabbing method for detection and quantification of HAV and noroviruses on food surfaces to enable quality assured analyses and PT distributions for this methodology.

Description:

The EURL has implemented a swabbing method according to ISO 15216 and distributed one qualitative PT distribution with bell pepper as matrix. Verification of the method is needed for accreditation and the quality assurance future PT distributions. Relevant performance criteria as LOD and LOQ will be determined.

In short, 10 samples with 10 different dilution levels will be analysed. Anticipated levels of viral genomes will be statistically analysed towards obtained results. A verification report will be written to be used for accreditation of the method.

An SOP has been written and shared with the NRLs. The SOP will be updated according to performance criteria obtained.

Expected Output:

An implemented and verified method for detection and quantification of HAV and norovirus on food surfaces.

Duration:

2023 and 2024

Sub-activity 3.1.4 Verification of method for detection of norovirus and hepatitis A virus in leafy green vegetables according to ISO 15216.

Objectives:

To verify the method for detection of norovirus and hepatitis A virus in leafy green vegetables towards performance criteria described in ISO 15216.

Description:

The method has previously been accredited for berries, but no corresponding accreditation for leafy vegetables has been done yet. A verification plan has been prepared and the verification will be done during 2023.

In the ISO 15216 protocol, the use of a miniMag® equipment from BioMérieux is indicated in the RNA-extraction step. BioMérieux will not produce this equipment anymore and instead will provide a device called eGENE-UP®. As several NRLs already have this equipment the EURL bought this device in order to evaluate PT samples with this alternative equipment. Initial tests indicated that the new equipment is interchangeable with the miniMAG®. Verification of the new device has been performed by the EURL in 2022 so the verification of leafy green vegetables will be executed using eGENE-UP®.

Expected Output:

An accredited ISO 15216 method for detection and quantification of HAV and norovirus in leafy green vegetables.

Duration:

2023

Sub-activity 3.1.5 *Implementation and verification of method for quantification of norovirus and HAV in bottled water.*

Objectives:

To implement and verify a method for quantification of norovirus and HAV in bottled water.

Description:

Questions has been raised by NRLs regarding methods for testing bottled waters for foodborne viruses and if it could be an option for PT distributions. The EURL will distribute bottled water as a PT in 2024. Prior to the PT, development and verification of the method for bottled water will be carried out. The EURL has previous experience of methods for quantifying viruses in water samples but not according to ISO 15216.

The ISO-method needs to be implemented and there is a need for verification of the method to determine performance characteristics as LOD and LOQ.

Expected Output:

An implemented and verified method for quantification of norovirus and HAV in bottled water to enable PT distributions for this methodology, 2023.

Duration:

2023 and 2024

Sub-activity 3.1.6 *Development of mengovirus RT-PCR for improving the stability of the results of analyses using ISO 15216 standard method.*

Objectives:

Improve methodology for relative quantification of the mengovirus, to reach a better stability of the process control used in ISO 15216.

Description:

A basic requirement for detecting viruses in food involves the use of a process control virus for monitoring the quality of the extraction procedure as described in the ISO 15216 standards. Mengovirus is used as a process control due to its similar morphological and physicochemical properties as norovirus and HAV. However, the current mengovirus RT-PCR gives occasionally instable results as experienced by the EURL and NRLs. The EURL will develop the RT-PCR by designing new primers and probe for mengovirus amplification. The method will be tested on matrix groups included in ISO 15216 prior to verification.

Expected Output:

1. Stabilised methods included in ISO 15216.
2. A developed and tested mengovirus RT-PCR that gives more stable results.
3. Verification of the developed method for RT-PCR.

Duration:

List of Indicators WP1:

- Number of laboratory methods for which details and guidance as regards their techniques, validation and interpretation are available in the EURL website
- Number of new laboratory methods developed in the reporting period
- Number of laboratory methods improved in the reporting period
- Number of Proficiency Tests (PTs) organised by the EURL for national reference laboratories/NRLs (indicate for each year – 2023 & 2024)
- Cost of PTs
- Number of Comparative Tests (CTs) organised by the EURL for NRLs (indicate for each year – 2023 & 2024)
- Cost of CTs
- Success rate of Member States NRLs/OLs in PTs/CTs and, if necessary, corrective action
- Number of corrective actions undertaken (aggregated data on corrective actions for all NRLs)

3.2. Work Package 2 – ASSISTANCE TO NRLs

Objectives: TO PROVIDE SCIENTIFIC AND TECHNICAL ASSISTANCE TO NRLs

- *Art. 94.2.d Coordinating practical arrangements necessary to apply new methods of laboratory analysis, testing or diagnosis, and informing national reference laboratories of advances in this field.*
-

- *Art. 94.2.e Conducting training courses for staff from national reference laboratories and, if needed, from other official laboratories, as well as of experts from third countries.*
-
- *Art. 94.2.g Providing information on relevant national, Union and international research activities to national reference laboratories.*

Sub-activity 3.2.1 Annual workshop

Objectives:

To inform NRLs about PT results, development of standardisation processes and scientific progress concerning foodborne viruses and the detection and quantification of those in food matrices. Furthermore, to discuss the actual needs among NRLs and future work that can be done in the network.

Description:

The 2021 workshop was held as a digital meeting and a physical workshop was held in Uppsala in June 2022. As requested by the NRLs, DG SANTE, EFSA and ECDC were invited to both workshops. Physical workshops are more valuable and effective compared to the digital meetings so the aim is to hold physical meetings both in 2023 and 2024. The budget will include two physical meetings, one each year. In 2023, the workshop will be held in Uppsala, Sweden. In 2024, the workshop will be hosted by the NRL at the Marine Institute on Ireland.

In general, the workshops will contain information on PT results, information about standardisation processes, updates on relevant legislation, and progress in method development. Furthermore, discussions will be held about how NRL network collaboration promotes scientific progress and how the network cooperation may facilitate harmonisation of analytical results amongst Member States.

More specific:

The discussion on microbiological criteria for norovirus in oysters will be discussed and possibilities for the network to support the Commission towards a relevant decision on the matter will be further evaluated.

A specific section for NGS techniques will be included at each workshop.

Presentations of participating NRLs on method development and surveillance is also included.

Expected Output:

1. A well informed NRL network about harmonisation and standardisation progresses.
2. An NRL network that shares information on method development for improved analysis of foodborne viruses in complex matrices.
3. An NRL network functioning as a facilitator of progresses in the scientific field of foodborne viruses with the aim to decrease public health burden of foodborne virus transmission.

Duration:

2023 and 2024

Sub-activity 3.2.2 *Technical training courses*

Objectives:

To give theoretical background and hands-on training in analytical methods and techniques to less experienced NRLs.

Description:

A webinar on verification of the ISO 15216 method as a basis for accreditation was held in 2021. The meeting contained discussions on specific laboratory and national demands to obtain accreditation. It served as a very good input to the guide for verification of ISO 15216 in single laboratories which will be published as an EURL Guidance document (see sub activity 3.2.3).

In autumn 2022 a course on the quantification of HAV and noroviruses in BMS was held on two occasions with in total 15 participants and 4 course leaders. The planned courses for 2023 and 2024 have been decided according to the requests of NRLs.

A technical course on norovirus and HAV quantification in soft fruits will be held in May 2023 prior to the PT distribution with the same matrix planned for September 2023.

A basic course in the use of NGS for typing of norovirus from food samples will be held in 2024.

Future PT distributions will give the possibility for the NRLs to provide results from both RT-qPCR and RT-dPCR. Therefore, a practical course on RT-dPCR will be held in autumn, 2024.

An Inter EURL WG on NGS joint course will be held in 2023, similar to the one given in Italy in 2022. Three participants from the NRL network for Foodborne Viruses will be offered participation and reimbursement

Expected Output:

1. Better method performance among the NRLs.
2. Support to the NRLs on NGS to be used for norovirus typing.

Duration:

2023 and 2024

Sub-activity 3.2.3 *Preparedness of staff and assistance to NRLs*

Objectives:

To follow the national and international scientific development in the area of foodborne viruses and to inform about the EURL and its activities. Support NRLs with scientific and methodological knowledge in a timely manner. The EURL website should be continuously updated and improved as a means of communication with NRLs and other stakeholders.

Description:

This activity seeks to keep the staff knowledgeable in the area of foodborne viruses to be able to support NRLs in the best way with such knowledge. Workshops and training courses organised by the EURL will be continuously evaluated to improve those activities over the years. Preparation of work programs and budget are included as well as the internal organisation of the EURL. During the last WP, the EURL experienced staff problems due to long term sick leaves. The organisation is now stronger, as we built up a more resilient organisation where staff are more able to replace each other for different tasks performed by the EURL. Further the current director is not getting younger and a replacement is prepared, probably during the WP for 2025 and 2026. New staff has been employed (Katalin Nemes) with long term experience in virology and is now building up specific experience in the field of foodborne viruses. In December 2022 one of the staff members (Sofia person) defended her thesis for a doctoral degree in the area of methodology for detection and quantification of food borne viruses in food. In the field of NGS one of the staff members (Ronnie Eriksson) presented his master's thesis during spring 2023, specifically dedicated to typing methods of norovirus using NGS technique.

It is important to follow scientific publications as well as to participate in conferences. Two staff members will participate in the biennial Food and Environmental Virology Conference in Tokyo during 2024. Relevant abstracts will be presented about progress in method development achieved at the EURL. The Swedish Society for Virology has a yearly symposium. One staff member will participate each year to strengthen the good contact with the Swedish virology community and the hospital laboratories on which we depend for the collection of reference materials, especially faecal samples used for contamination of food matrices in method development, validation studies and PT distributions.

During this WP, the EURL plan to make a study visit to the Swedish control of BMS production areas on the West Coast of Sweden. The idea is to improve the staff members' knowledge and understanding of the control and production of BMS. During two days six member will be guided by the control unit at the Swedish Food Agency.

During previous workshops there has been a request to provide a guidance document on verification towards the validated ISO method ISO 15216 to facilitate the accreditation process for NRLs and other official laboratories in the Member States. The guide has been shared with the NRLs and will be published as an EURL Guidance Document in 2023. An R programming language based application has been developed to facilitate the calculation of parameters such as LOD and LOQ. The program will be published in connection with the publication of the guide.

The NRLs have requested bioinformatics surveillance of the inclusivity of norovirus and HAV primers and probes recommended in ISO 15216. This surveillance aims to show if new sequence variants of these viruses published in GeneBank, NoroNet and HAVnet are indicated to not be detected with current primers and probes and therefore require new primer- and/or probe design. This work will be performed by the EURL on a biannual basis in the future

Expected Output:

1. Improved competence among and organisation of staff members to enable improved support to the NRLs.
2. Opportunities to inform the scientific society about the EURL activities.
3. The publication of an EURL Guidance Document for the verification of ISO15216, to facilitate NRL accreditation processes.

Duration:

2023 and 2024

List of Indicators WP2:

- Number of enquires requested from national support networks and bodies and competent authorities
- Number and quality of replies provided to enquiries of national support networks and bodies and competent authorities
- Number of training and collaboration activities with national support networks and bodies and competent authorities
- Number of workshops & meetings organised
- Cost for workshops & meetings and average cost for participant
- Number of technical assistance provided by the EURL to NRLs enquiries

3.3. Work Package 3 – ASSISTANCE TO THE EUROPEAN COMMISSION AND OTHER ORGANISATIONS

Objectives: TO PROVIDE SCIENTIFIC AND TECHNICAL ASSISTANCE TO THE EUROPEAN COMMISSION AND OTHER ORGANISATIONS

- *Art. 94.2.f Providing scientific and technical assistance to the Commission within the scope of their mission.*
- *Art. 94.2.h Collaborating within the scope of their mission with laboratories in third countries and with the European Food Safety Authority (EFSA), the European Medicines Agency (EMA) and the European Centre for Disease Prevention and Control (ECDC).*
- *Art. 94.2.i Assisting actively in the diagnosis of outbreaks in Member States of foodborne, zoonotic or animal diseases, or of pests of plants, by carrying out confirmatory diagnosis, characterisation and taxonomic or epizootic studies on pathogen isolates or pest specimens.*

Sub-activity 3.3.1 *Participation in the EURL Working Group for NGS.*

Objectives:

Further promotion of the usage of next generation sequencing (NGS), across the EURLs' network and method development

Description:

Beside the EURL working group for NGS, the EURL for Foodborne Viruses have formed a WG for NRLs for Foodborne Viruses on NGS. The goal for the WG is to increase the possibility for a faster development and harmonization of NGS techniques used in the area of foodborne viruses and to promote collaboration and knowledge transfer between laboratories. The group have physical or digital meetings in connection to the yearly workshop. The WG is now transformed into a core group of experienced NRLs to enable faster progress in the area. The NRLs agreed to this change on the 2022 workshop and the decision was included in the resolutions. Prior to the yearly workshop there is an organised meeting for the NGS Core Group. The 2023 EURL workshop will be in Uppsala and the NGS core group meeting at the Swedish Food Agency. General discussions on NGS will now be included in the yearly workshop. Production of a workflow document for NGS for typing of norovirus will be part of the assignments of the Core Group. This will also be used as an input to the inter EURL WG on NGS.

The group organized a course at the NRL at IFRAMER, France in the basic use of NGS for typing of norovirus during late 2022, which was much appreciated but could not harbour all the applicants. A new course will be planned during 2024 with the help of the Core Group.

The EURL will participate in two meetings in the Inter EURL WG on NGS each year and give continuous input to the EURL working group.

Expected Output:

Support to the commission, EFSA and ECDC in their work to ensure the development of NGS as a typing method in EU MS.

Duration:

2023 and 2024

Sub-activity 3.3.2 *Scientific assistance and emergency support to the Commission, EU agencies and third countries.*

Objectives:

To ensure that the EURL staff is well trained, up-dated and knowledgeable in the area of foodborne viruses, so that appropriate expertise can be provided to the Commission and that adequate support will be given in emergency situations, for example during foodborne viral outbreaks.

Description:

Sub-activity 2.3 will support this activity and requests from the Commission and EU agencies for scientific and technical assistance will have priority and be handled by the EURL scientific staff in a timely manner.

Expected Output:

A well informed and trained staff that can give support to the Commission and EU agencies in a timely manner.

Duration:

2023 and 2024

Sub-activity 3.3.3 International standardisation of relevant methods in the area of foodborne viruses.

Objectives:

To ensure the availability of standardized methods, for the detection and quantification of hepatitis E virus (HEV) in animal products and standardised methods for the validation of new methods against a reference method, as a basis for harmonisation in the NRL network and official control in EU MS.

Description:

The ISO standard 16140 are under revision and concerns an update of the verification and validation of methods for the detection and quantification of microorganisms in food and feed. A new part of ISO 16140 is under development to be used for viruses and parasites. This is especially relevant for the ISO 15216 method.

Cases of hepatitis E have drastically increased in most EU MS during the last 15 years. The seroprevalence varies between countries and regions but are generally around 10 %, but in areas with specific eating habits, over 50 % has been recorded.

In 2017 EFSA published a scientific opinion on HEV. The first conclusion states “The validation and standardisation of methods for detection and quantification of HEV from meat and meat products should be a high priority”.

ISO/TC34/SC9 has formed a working group to standardise methods for the detection of HEV in animal products. The WG is led by Nigel Cook (UK). The outlined work plan is currently followed and methods to include are stated.

The EURL will continue its participation in the HEV working group.

The EURL budget includes participation in ISO/TC34/SC9 relevant working groups and a limited amount of working days is included in the budget.

The EURL is represented through the Swedish Institute for Standards and should in the future be represented as an EU organisation.

Expected Output:

1. A standardised procedure for the validation of methods concerning viruses and parasites.

2. A standardised method for the detection and quantification of hepatitis E virus in animal products.
3. In the long run a harmonisation process of the detection of HEV in the NRL network and thereby national official control laboratories.
4. Ultimately, reduced disease burden through future management measures.

Duration:

2023 and 2024

Sub-activity 3.3.4 Supporting document for the decision on microbiological criteria for norovirus in oysters consumed raw.

Objectives:

To give scientific support to the Commission to enable an informed decision on microbiological criteria for noroviruses in oysters.

Description:

To increase the possibility to make a rational decision when it comes to the outline of microbiological criteria for norovirus in oysters, the EURL will provide two supporting documents to the Commission. The EURL will compile outbreak data from MS through the NRLs and compile knowledge from the scientific literature to understand the dose response relationship of norovirus levels in oysters and the risk for disease.

The ISO 15216-1 is the basis for quantification of norovirus in oysters. The EURL will, in a supporting document describe the performance criteria of ISO 15216-1 to facilitate a rational decision on the subject.

Working days are included in the budget.

Expected Output:

Two documents to support the Commission and Member States in their future decision on microbiological criteria for norovirus in oysters consumed raw.

Duration:

2023 spring 2024

List of Indicators WP3:

- Number of technical and scientific feedback provided by the EURL based on European Commission enquiries
- Number of collaboration activities with other organisations

3.4. Work Package 4 - REAGENTS AND REFERENCE COLLECTIONS

Objectives:

- *Art. 94.2.j Coordinating or performing tests for the verification of the quality of reagents and lots of reagents used for the diagnosis of foodborne, zoonotic or animal diseases and pests of plants.*

- *Art. 94.2.k Where relevant for their area of competence, establishing and maintaining:*
 - i. reference collections of pests of plants and/or reference strains of pathogenic agents;*
 - ii. reference collections of materials intended to come into contact with food used to calibrate analytical equipment and provide samples thereof to national reference laboratories;*
 - iii. up-to-date lists of available reference substances and reagents and of manufacturers and suppliers of such substances and reagents.*

Sub-activity 3.4.1 Production of standard DNA, control RNA and process control virus

Objectives:

To ensure that NRLs have access to standards and controls to increase uniformity of method performance between laboratories. To provide means for quality assurance and method evaluations to laboratories.

Description:

DNA standards and control RNA as well as process control virus (mengovirus) recommended in ISO 15216 will be produced and included in PT distributions.

The EURL will evaluate so called gBlocks as an alternative standard for the quantification of noroviruses and HAV. gBlocks Gene Fragments are double-stranded DNA fragments designed for affordable and easy gene construction or modification, suited for qPCR and NGS controls. Each gBlocks Gene Fragment goes through a quality control process, which includes size verification by capillary electrophoresis and sequence identification by mass spectrometry. This rigorous testing could potentially decrease differences between laboratory results. gBlocks are commercially available and could easily be uniformly produced and the exact same standard could be used of all laboratories involved in quantification of noroviruses and HAV according to ISO 15216. gBlocks can be designed to facilitate the production of control RNA used in ISO 15216. A preliminary test by the EURL indicated that gBlocks work well to generate stable standard curves. Implementation and evaluation will be carried out by the EURL during 2023 and 2024

Expected Output:

1. Production of standards and controls to increase uniformity of method performance.
2. Overall improved quality assurance of the analyses performed at the NRLs for detection of foodborne viruses.

Duration:

2023 and 2024

Sub-activity 3.4.2 *Organise and collect reference materials such as faecal samples and cultivated viruses.*

Objectives:

To have an up-to-date collection of reference substances and reagents and to store these in an accessible manner.

Description:

Continuous collection of reference material as norovirus positive faecal samples has been fairly successful during 2022 and the EURL will probably have the possibility to share material with NRLs during 2024. Norovirus positive faecal material will be characterized and quantified during 2023.

In cooperation with hospitals and other partners we will continuously collect norovirus faecal samples and other relevant enteric viruses to be registered and stored. Typing of such virus stocks will be done.

Mengovirus will be cultivated in cooperation with the Swedish National Veterinary Institute. Costs for this is included in the budget.

Expected Output:

A collection of stored and available substances and reagents is maintained and continuously updated. Relevant reference materials may be provided to NRLs and other stakeholders.

Duration:

2023 and 2024

3.5. REQUIREMENTS RELATED TO OTHER LEGISLATION

Not applicable.

4. REMARKS

(if applicable)