Public Health England

Protecting and improving the nation's health

2019 to 2020 Report of UK National Reference Laboratory for Food Microbiology

Activities for *Listeria monocytogenes*, coagulase positive staphylococci, *Escherichia coli* (including STEC), *Campylobacter*, *Salmonella* and antimicrobial resistance

April 2019 to March 2020

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Executive summary

The UK's National Reference Laboratory (NRL) for food microbiology is provided by Public Health England for the Food Standards Agency (FSA), as part of the UKs compliance to the Regulations (EU) 882/2004 and 2017/625 for Official Control Regulations (OCLs) for food safety. This is the annual report of the NRL's activities between April 2019 and March 2020 and relates to activities for *Listeria monocytogenes*, coagulase-positive staphylococci, *Escherichia coli* (incl. STEC), *Campylobacter*, *Salmonella* and antimicrobial resistance (AR).

NRL quarterly newsletters and European Reference Laboratory (EURL) information were disseminated to the FSA, the Official Control Laboratories (OCLs) and other stakeholders. An annual OCL user day was held to inform OCLs of information from the EURLs, legislation changes and methodology updates. The third OCL audit was sent to OCLs in October 2019 and reveal varying testing capabilities and Food Examiner (FE) availability. The NRL also advised and contributed of their position regarding EU Exit and the new OCRs to other departments in PHE, DEFRA and FSA.

All 6 EURL meetings were represented either as the UK NRL or in an individual expert capacity, and impartial advice was provided to FSA, OCLs and other laboratories throughout the year. The NRL also attended 3 EURL training events and found them extremely useful. The NRL was active in the BSI AW9 microbiology committee and has continued to be a member of the CEN TAG18 expert working group for the revision of the ISO TS 13136 (PCR detection of shiga toxin-producing *Escherichia coli*).

There are 11 relevant methods provided by the NRL to OCLs on the .gov.uk website. Other methods not on the .gov.uk website are available upon request.

Thirteen OCLs registered to the European Food Microbiology Legislation (EFL) External Quality Assessment Scheme, under NRL support for 2019 to 2020. Overall, results were satisfactory, however, this year included tests from the Microbiological Criteria that are infrequently performed or recently added in the UK. This led to a lower performance and OCLs were advised on improvements in these areas and to adhere to reporting instructions for the EFL scheme. The NRL participated in 10 EURL PTs and there was satisfactory performance for 9 of the 10 distributions. Two Skype workshops were organised for OCLs; 'Implementation of ISO 17025:2017' and 'Principles of PCR and Real-time PCR in Food Microbiology'. Feedback was good overall for both events. Future topics for training will be based on OCL requests.

Details of the proposed NRL activities for 2020 to 2021, and a timeline to achieve these complete this report.

Abbreviation list

AFBI	Agri-Food and Biosciences Institute (Northern Ireland)
APHA	Animal and Plant Health Agency
AR	Antimicrobial resistance
BSI	British Standards Institute
CEFAS	Centre for Environment, Fisheries and Aquaculture Science
CEN	European Committee for Standardization
COVID-19	Coronavirus disease 2019
CPS	Coagulase-positive Staphylococci
DEFRA	Department for Environment, Food and Rural Affairs
ECDC	European Centre for Disease Prevention and Control
EFL	European Food Microbiology Legislation
EFSA	European Food Safety Authority
EPIS	Epidemic Intelligence Information System
EQA	External Quality Assurance
ESBL	Extended-spectrum Beta-Lactamases
EURL	European Reference Laboratory
FAO	Food and Agriculture Organization of the United Nations
FBO	Food Business Operator
FE	Food Examiner
FEPTU	Food and Environmental Proficiency Testing Unit
FSA	Food Standards Agency
FSS	Food Standards Scotland
FW&E	Food, Water and Environment
ISO	International Standards Organisation
JEMRA	Joint FAO/WHO Expert Meetings on Microbiological Risk Assessment
MIC	Minimum inhibitory concentration
MLST	Multi-locus sequence typing
MRSA	methicillin-resistant *Staphylococcus aureus*
MS	Member State
mTSB	modified soy broth
NRL	National Reference Laboratory
OCL	Official Control Laboratory
OCR	Official Control Regulations
PCR	Polymerase Chain Reaction
PFGE	Pulsed-field Gel Electrophoresis
PHE	Public Health England
PT	Proficiency test
SOP	Standard operating procedure
STEC	Shiga-toxin producing *E. coli*
UoM	Uncertainty of Measurement
WG	Working Group
WGS	Whole Genome Sequencing
WHO	World Health Organization

Introduction

The EU Regulation 2017/625 (previously (EU) 882/2004) for Official Controls requires each Member State to have a National Reference Laboratory (NRL) to be responsible for different aspects for *Listeria monocytogenes*, coagulase-positive staphylococci, *Escherichia coli* (incl. STEC), *Campylobacter*, *Salmonella* and AMR. Public Health England (PHE) has provided this service as the UK's NRL for food microbiology since 2011 under a contract with the UK's Competent Authority, the Food Standards Agency (FSA). This and other EU regulations will be transferred into UK Statutory Instruments once the UK leaves the EU; therefore, the NRL has been instructed by FSA to continue these activities until March 2023.

This report details the secretariat services, advice and representation within the UK/EU, production of documents, coordinating and participating in audits, ring trials and European Reference Laboratories (EURLs) initiatives and communication of results and data between April 2019 and March 2020. Table 1 list the NRL core functions and the activities are described in this annual report.

Table 1. PHE NRL Core Functions, April 2019 to March 2020

Core function	Description
1	Secretariat services
1.a	Disseminate information/advice from international organisations to FSA, OCLs and other UK laboratories in a timely and effective manner
1.a	Produce and circulate quarterly newsletters to FSA, OCLs and other UK laboratories
1.a	Co-ordinate the OCL User Day to update UK OCLs and other relevant UK laboratories to the NRL core functions
1.a	Assist in the dissemination and advice of EPIS and other alerts from the appropriate authority
1.b	Update and perform an audit to gather information regarding the OCL's capabilities and requirements
1.b	Review content of the UK Food Examiner Register
1.b	Continue liaison meetings with APHA for AR, *Campylobacter* and *Salmonella*
1.c	Liaise with FSA in matters arising from implementing the new Official Control Regulation, (EU) 2017/625
1.d	Provide regular updates to the FSA on NRL activities by producing monthly reports and meet on a quarterly basis
1.d	Produce and submit annual report to the FSA on NRL activities for 2019 to 2020
1.e	Maintain and update the NRL web content on the PHE website

2	Advice and representation within the UK and internationally
2.a	Provide impartial expert advice to FSA, OCLs and other UK laboratories, upon request
2.b	Represent the UK at relevant international meetings and working groups; consult FSA prior to meetings and submit an internal report after attendance of meetings
2.c	Attend training workshop at international organisations, where relevant and after successful applications
2.e	Keep abreast of methodology developments and advise FSA and OCLs (for example, workflow and Service Level Agreement for CPS toxin testing)
2.g	Participate in the BSI AW9 microbiology committee
2.g	Participate in Working Group to revise the ISO/TS 13136:2012 (PCR detection of STEC)
3	Production of standard operating procedures, codes of practice and guidance documents
3.a	Update and expand food methods archive on NRL website
3.a	Prepare a guidance document for OCLs and the FSA on the use and validation of alternative methods for testing Official Controls
3.a	Produce a poor performance protocol for OCL participation in the EFL proficiency test scheme
3.a	Draft a manuscript for peer-review summarising 4 years of OCL performance of the EFL PT scheme
4	Compliance assessment via audits and ring trials
4.a	Ensure consistency and quality of testing approached applied by UK OCLs and support where necessary
4.b	Liaise with FEPTU and monitor OCL's comparative testing performance and assist OCLs in the implementation of corrective measures
4.b	Evaluate OCL's performance using trend analysis
4.c	Coordinate the participation of OCLs in international method validation studies and other initiatives and report to FSA
4.d	Participate as UK-NRL in ring trials including method validation studies organised by the EURL (where available) and report to FSA
4.e	Organise a Skype/classroom-based workshop for UK OCLs on the implementation of ISO 17025:2017
4.e	Organise a practical workshop for UK OCLs, dependent on the outcome of the audit (1.b) and OCL needs
5	Co-ordination within the UK of international initiatives
5.a	Support the food aspect of the EU-wide AR monitoring (Decision 2013/652/EU), liaising with FSA, OCLs relevant Reference Laboratories and APHA.
5.a	Liaise with APHA, audit and review strategy for harmonization of existing antimicrobial resistance testing
5.a	Provide information regarding Whole Genome Sequencing when requested from the EURLs, and participate in any related workshops, training and guidance documentation, where necessary
6	Communication of results and data use

Core Function One: Secretariat services

Dissemination of information from the EURLs

The 6 EURLs send information concerning new reports, outbreaks and other related topics to the NRL. Information is then cascaded to the appropriate personnel and stakeholder(s) (e.g., OCLs, FSA, PHE Epidemiology and Reference Units, Scottish Reference Laboratories, APHA, AFBI and CEFAS), with any additional information or advice on further steps to be taken. The NRL also receives questionnaires and surveys from the EURLs regarding NRL and/or country-wide practices. These communications are described below by work activity with links to the EURLs' websites; information concerning meetings, training, proficiency tests (PTs) and ISO standards are incorporated in to the relevant sections of this report. Where available, the EURLs' work programmes can be found in the Annex.

Listeria monocytogenes

EURL Website: https://eurl-listeria.anses.fr/en/minisite/listeria-monocytogenes/eurl-listeria-monocytogenes

From April 2019 to March 2020, the NRL received 5 reports (3 produced by the EURL, 2 by EFSA/ECDC), news items and the launch of an updated website (see above link).The *Listeria* EURL also circulated various resources available for NRLs, including a WGS pipeline developed by Anses, certified reference material for PFGE analysis and educational slides developed by the EURL shelf-life working group, targeting Competent Authorities in the various aspects of shelf-life and challenge testing.

There have been 2 Epidemic Intelligence Information System (EPIS) enquiries of *L. monocytogenes* clusters sent from the *Listeria* EURL for this reporting period. The NRL log these and contacts the relevant staff in PHE to respond back to the EURL with the requested information.

In May 2019, the EURL sent a request from EFSA to announce to MSs that the European Molecular Typing Database is ready to receive data, and for participants to register and sign the Collaborative Agreement.

In July 2019, the EURL enquired whether any MSs had data on the temperatures of domestic refrigerators, to further refine data within the EURL Technical Guidance for shelf-life testing. The NRL responded to inform the EURL that the UK does not have this type of data.

Coagulase-positive staphylococci

Website: https://eurl-staphylococci.anses.fr/en/minisite/staphylococci/about-coagulase-positive-staphylococci

The CPS EURL notified the NRLs of an updated website with new posts during this reporting period and e-mailed a tailored report regarding CPS methodology from the ISO plenary meeting for methods applicable to the food chain.

The NRLs received a link from the EURL of their recommended method to detect the staphylococcal toxin genes by real-time PCR, which was circulated to relevant UK colleagues. A guidance on how to implement ISO 6888-1 and 2 standard (enumeration of CPS in foods) in accordance to ISO 7218 standard (general guidance to microbiological examinations) was also circulated to the NRL network by the EURL.

Escherichia coli (including STEC)

Website: http://old.iss.it/vtec/

The NRL received 14 outbreak alerts from the EURL; 9 from USA, and 5 from individual European countries. The EURL also sent an announcement in May 2019 for the 1st International Conference of the European College of Veterinary Microbiology, which took place on 26-27 September 2019 and was sent to APHA.

The EURL sent 2 external reports in this reporting period; The World Food and Agriculture Statistical Pocketbook 2019 from the Food and Agriculture Organization of the United Nations (FAO) and an EFSA Scientific Opinion on the ‘Pathogenicity assessment of Shiga toxin-producing *Escherichia coli* (STEC) and the public health risk posed by contamination of food with STEC’.

In September 2019, the EURL passed on a request from FAO/WHO to seek experts on control of STEC in meat and dairy products to join the Joint FAO/WHO Meetings on Microbiological Risk Assessment (JEMRA) meetings to give scientific advice on microbial food safety issues; this was forwarded to relevant UK colleagues.

The EURL requested all NRLs to check the contact details on the EURL website and also cascaded their planned activities for 2020.

Campylobacter

Website: http://www.sva.se/en/service-and-products/eurl-campylobacter

In July 2019, the EURL sent a short questionnaire to all NRLs concerning the implementation of the Process Hygiene Criteria (PHC) in their country, including sampling and methodological aspects. This was sent to FSA to further refine the answers and the collated information was submitted before the deadline.

The EURL also circulated news of future events to all the NRLs, including a training course based on the ISO 10272:2017 series, the NRL network workshop in September 2020, and 3 PTs to be sent in March 2020.

In November 2019, the NRL received an email requesting to submit *Campylobacter* manuscripts to a special issue of the journal Pathogens, to which former EURL colleagues were guest editors. This was forwarded to relevant *Campylobacter* experts.

Salmonella

Website: https://www.eurlsalmonella.eu/

Four quarterly EURL newsletters were received by email, informing NRLs of EURL activities, such as proficiency tests, workshop preparation and literature searches of relevant *Salmonella* scientific papers. These were forwarded to the FSA, the OCLs and other relevant laboratories in the UK. The EURL newsletters can be found in the Annex.

There was EURL correspondences concerning 5 Epidemic Intelligence Information System (EPIS) *Salmonella* clusters between April 2019 and March 2019. These are processed in the same way as the *Listeria* enquiries, see above.

The EURL also wrote to NRLs in March 2020 to request whether the *Salmonella* testing on poultry before slaughter are still being performed under Regulation (EU) No. 200/2012, as COVID-19 were causing many European countries to lockdown and were experiencing staff shortages.

Antimicrobial resistance

Website: https://www.eurl-ar.eu/

The EURL forwarded external reports regarding antimicrobial resistance over the reporting period to the NRLs, including the joint EFSA and ECDC EU report on antimicrobial resistance in zoonotic and indicator bacteria from humans, animals and food in 2018, the WHO report of critically important antimicrobials in humans and others from Europe, USA and Canada. The EURL sent an external survey regarding AR in animals, which is not relevant to the NRL.

In September 2019, the EURL circulated a publication led from the EURL reviewing available tools to track global antimicrobial resistance (Hendriksen *et al*, Frontiers in Public Health, 2019) to the NRLs. This was circulated to relevant colleagues, as well as other publications.

The EURL sent their annual newsletter to all NRLs in December 2019 (see Annex), detailing new minimum inhibitory concentration (MIC) panels for the upcoming legislation, using ResFinder 4.0 for *in silico* antibiograms and other activities in the EU. They also circulated an external newsletter from EARS-Vet, which is funded by the EU Health Programme and 2 AR individual opportunities from different organisations.

The EURL announced in February 2020 the availability of 2 updated methods on their website for isolating ESBL, ampC and carbapenemase-producing *E.coli* from fresh meat and caecal specimens.

The EURL sent an urgent enquiry on behalf of ECDC and EFSA in January 2020 for NRLs to share any WGS data on OXA-244 carbapenemase-producing *E.coli* from food, feed or animals to support an outbreak investigation. This was the first such enquiry from the AR EURL and was logged and processed in the same way as EPIS enquires.

Parallel correspondence from various EURLs

EURLs are involved in joint horizontal activities and regulations; consequently, the UK NRL receives the same information from different EURLs. Between April 2019 and March 2020, the following were received and cascaded to relevant stakeholders:

- a joint EURL ‘Guidance Document for the organisation of Proficiency Tests by NRLs for national networks, including partial outsourcing’
- the EU One Health 2018 Zoonoses report
- a conference invitation ‘Modern technologies to enable response to crises: Next Generation Sequencing to tackle food-borne diseases in the EU’ which was due to be held on 10 March, but has been postponed to 25 September due to COVID-19
- invitation to joint EURL training on tools used in WGS (Bioinformatics for analysis of WGS, July 2019)

Related to Core Function(s): 1.a, 1.c, 2.d, 2.e, 2.f, 4.c, 5.a

Production of NRL quarterly newsletters

Quarterly newsletters have been produced by the NRL since 2016, and these allow OCLs and other stakeholders to gain information on NRL activities and areas that could affect them. A brief description of the newsletters' content is listed below and are available in the Annex:

- June 2019 covered the 2019 OCL User Day, a Skype session to implement ISO 17025:2017, proficiency testing from the EURLs and the EFL scheme and a brief method update
- September 2019 included the implementation of the new Official Control Regulations (OCRs), the launch of the OCL audit and a NRL-led uncertainty of measurement Skype session
- December 2019 reported of the OCRs coming into force on 14 December, a digest of the EU One Health 2018 Zoonoses Report, a methods update and upcoming relevant events
- March 2020 on how EU Exit and COVID-19 has affected attendance to EU meetings, an NRL-led principles of PCR Skype session, a surge of available and relevant documents and the temporary suspension of FEPTU's services, including the EFL PT scheme because of Covid-19.

Related to Core Function(s): 1.a, 2.d, 2.e, 2.f

Co-ordination of the 2019 OCL User Day

The seventh Official Control Laboratories User Day was held at PHE Colindale on 18 June 2019, where 32 colleagues attended from 11 OCLs, the PHE's Gastrointestinal Bacteria Reference Unit (GBRU) and the Food and Environmental Proficiency Testing Unit (FEPTU), Animal and Plant Health Agency (APHA), FSA, FSS and Campden BRI as well as all staff from the NRL.

The format of the day was updated this year, from having separate reports from each of the 6 EU Reference Laboratories (EURLs), to combining aspects of their shared activities, such as European epidemiology and NRL participation of EURL PTs, as single presentations. This was to reduce repetition, increase engagement and allowed more questions to be asked from delegates. Talks were also presented on EU Exit and the impact it has had from colleagues from FSA, the London Port Health Authority and PHE. Additional presentations were on *Campylobacter*, *Listeria* and PHE's move to Whole Genome Sequencing to detect and characterise foodborne bacteria. (see Annex for programme).

Overall, feedback was very good from those attended. Slide presentations are available on request.

Related to Core Function(s): 1.a, 1.b, 1.c, 2.a, 2.d, 2.e, 2.f

Assist in disseminating and responding to EPIS and other alerts

Since April 2018, the NRL have been receiving EPIS and other alerts from the EURLs. These have been from the *Listeria* (11 in total) and *Salmonella* (11 in total) EURLs, and more recently 1 from the AR EURL.

The EPIS and other alerts are logged by the NRL and relevant staff in PHE are contacted, as the EURL are requesting information on any related isolates from non-human origin. The specialist PHE staff then looks in the UK database to identify any closely related strains relating to the cluster concerned. The NRL then responds to the EURL of any information from PHE colleagues concerning the enquiry.
Related to Core Function(s): 1.a, 2.a

2019 Audit – Official Control Laboratories' capabilities and requirements

The UK NRL has undertaken 2 audits since 2013 to ascertain the Official Control Laboratories' capabilities and range of microbiological testing for food. These audits have helped identify training and educational gaps, which the NRL organised and delivered to the OCLs. With recent updates in ISO Standards and the new EU Official Control Regulations (EU 2017/625), a third audit was launched in October 2019 to all UK OCLs.

The information and questions were updated using the online tool SelectSurvey (see Annex for questions). OCLs were sent the SelectSurvey link and it was completed by all the OCLs. Preliminary analyses have revealed:

- all 14 OCLs responded; 6 in England, 4 in Scotland, 3 in Wales and 1 in Northern Ireland
  - 12/14 OCLs are now UK Accreditation Service (UKAS) accredited under the new ISO 17025:2017
- testing is performed for local government (N=14 OCLs), port health authorities (9), FBOs (12), surveillance purposes (11) and for other purposes (4)
- there are between 1 and 7 food examiners in each OCL
  - at the time of the audit, there were 14 individuals undertaking Food Examiner training in UK OCLs
- 7/14 OCLs perform challenge or shelf-life testing for FBOs or other customers
- compared to the 2016 audit, the OCLs are practising differences in testing against the microbiological criteria
  - OCLs have changed some of their testing capability, mostly linked to the matrices tested
- concerning *Campylobacter* testing as the most recent addition to the Process Hygiene Criteria:

- only 4/14 are accredited for enumeration of *Campylobacter*
- a further 2 OCLs perform enumeration but are not accredited
- this is the same as 2016 and there has been no change in the UK's capability
- 4 OCLs would like the NRL to cover *Campylobacter* enumeration for a future training event

The data will be fully analysed in summer 2020 and a report published by the end of the year.
Related to Core Function(s): 1.b, 2.d, 2.e, 4.a

Review content of the UK Food Examiner register

The NRL has established and maintained a Food Examiner register since 2014 to assist FSA to rapidly contact the appropriate local support from the OCLs if they receive enquiries concerning microbiological testing/investigation of food. An update of information was gathered during the 2019 OCL audit and the register will be revised in 2020.
Related to Core Function(s): 1.b, 4.a

Liaise with APHA regarding mutual NRL activities (*Campylobacter*, *Salmonella* and antimicrobial resistance)

Due to the shared responsibilities for *Salmonella*, *Campylobacter* and antimicrobial resistance (AR), the designated microbiology NRLs from PHE, APHA and Agri-Food and Biosciences Institute (AFBI) for Northern Ireland have held 6-monthly liaison meetings since 2013. Given that the EURLs will only financially support a single NRL per Member State at annual workshops, training events and participation in ring trials, PHE and APHA agreed that EURL funding would be allocated on an alternating basis. However, if activities of one organisation take precedence, for example that APHA undertakes the current statutory AR testing in the food chain across the EU, then that NRL will take priority for EURL funding. These meetings therefore allow transparency between the 2 NRLs and to ensure there is co-ordination of activities.

Since the UK withdrew from the EU on 31 January 2020, the European Commission is treating the UK as a third country and continues to allow the UK to participate in the NRL networks. For this reporting period, the PHE NRL arranged 2 meetings via teleconference with APHA and AFBI in July 2019 and January 2020, where shared activities were agreed.
Related to Core Function(s): 1.b, 4.c, 5.a

Liaise with FSA regarding the new Official Control Regulation, (EU) 2017/625

A European Heads of Food Safety Agencies working group produced an interpretation document to clarify ambiguous laboratory aspects of the EC Official Control Regulation (OCR) 2017/625. This was sent from the FSA to the NRL in May 2019 and was further circulated to all the OCLs. Due to EU Exit, the NRL circulated the transferred OCR as a UK Statutory Instrument (SI) document UK SI 2019 No. 665 in March 2019 to the OCLs via the March newsletter. The FSA also held a public consultation on the implementation of the OCRs in August 2019, which the NRL sent comments directly to FSA.

Once the relevant OCR Articles concerning the OCLs came into force on 14 December 2019, Defra and FSA issued a factsheet to OCLs of the major changes from the old OCR (EC) No. 882/2004, and the OCLs' obligations to comply with the new OCRs. The NRL has been further consulted on a draft clarification document for the OCLs in March 2020 and have sent comments in April 2020.
Related to Core Function(s): 1.c, 2.a

Provide regular updates to Food Standards Agency

NRL representatives met with FSA quarterly (24 June 2019, 17 September 2019, 4 December 2019, via Skype 30 March 2020) to discuss progress made, difficulties met, and future or new activities (see Annex for minutes). In addition, monthly reports listing NRL activities have been submitted electronically to the FSA (see Annex).
Related to Core Function: 1.d

NRL Web Content

Since 2013, information about the NRL has been held on the Public Health England section of the GOV.UK website. There is access to NRL annual reports since 2013, 10 standard methods, a public health management guidance, and reports of the 2016 and 2013 OCL audits. There is also general information about the NRL, expert witness information, and contact details on the web page. The standard methods are periodically reviewed and updated, and the NRL will continue to increase the collection, where relevant.

The website address is https://www.gov.uk/government/collections/uk-national-reference-laboratory-for-food-microbiology . For ease of access, OCLs and other stakeholders are advised to use a search engine and type 'fwe nrl' or 'food NRL', as the NRL web page is normally the top hit.
Related to Core Function(s): 1.a, 1.b, 1.e, 3.a

Core Function Two: Advice and representation within the UK/EU

Provide impartial advice to FSA, OCLs and other UK laboratories

The NRL received requests for expert advice from small business organisations to European institutes.

General

Those between April 2019 and March 2020 are briefly reported and categorised as:

- from the EC, looking for a suitable laboratory to test 3 PFGE certified reference materials; the UK NRL no longer performs PFGE and was unable to help
- three separate requests to microbiologically test food, which was forwarded to the relevant laboratories
- a private company requesting *E.coli* isolates to test their laboratory assay
- a invitation to speak at the Chartered Institute of Environmental Health (CIEH) Food Safety Conference in June, which was accepted
- one student requesting a placement in a food laboratory; this was discussed within the PHE FW&E Microbiology Services.
- three laboratory enquiries concerning methodology; interpreting results from environmental swabs, an international request for an updated method and implementing PCR into an OCL laboratory; all of these were dealt by NRL experts
- following from a Skype session on Uncertainty of Measurement, 3 OCLs requesting further clarification and assurance of how to use the PHE spreadsheet
- an Environmental Health Officer (EHO) requesting information of how to extend the shelf-life of a product; the NRL advised to contact a couple of external organisations for expert advice and possible testing
- queries concerning disinfectants; a company offering their system of decontamination, FSA asking which current disinfectant guidance are available to FBOs, and a call for experts to join a CEN group for disinfectants
- an EU NRL requesting how the UK NRL for Food Microbiology distributes and formats the quarterly newsletters; the NRL gave advice and an example newsletter
- two COVID-19 related queries; via APHA of whether haulage drivers have the right of passage when carrying feed to farms: this was directed to the PHE COVID guidance cell and; a consumer asking whether freezer to plate meals were safe to store in the fridge and consumed by immunocompromised people: this was directed to FSA

Following discussions which commenced in August 2018, the UK NRL for Food Microbiology attended at least 3 PHE meetings to address further how the NRL activities would be affected by a no deal EU Exit and what contingencies are in place. From 1 September 2019 the FSA requested that the UK NRLs do not attend any EU meetings, after a UK governmental statement was published on 20 August https://www.gov.uk/government/news/uk-officials-will-stop-attending-most-eu-meetings-from-1-september . The PHE EU Exit office was informed of this and there was an agreement in place between FSA and PHE that individual experts could attend the EURL meetings, as these are of public health and health security importance for the UK. Consequently, the PHE EU Exit office requested details of all EU meetings that NRL and other reference laboratories were due to attend after 1 September 2019.

The UK NRL for Food Microbiology have contributed to the FSA review of Official Control Laboratories, which was coordinated in 2 phases and reported in September 2019.

Listeria

In April 2019, the *Listeria* EURL requested the UK to share any training documents or slides for challenge testing, to enhance their own training pack; the NRL sent material in the form of Powerpoint slides used for training.

A direct EFSA request for UK data of *Listeria* in frozen foods, following a UK survey which was partially presented at the *Listeria* EURL meeting, 10 to 12 April 2019; the UK provided data, which supported the recent EFSA ‘Scientific Opinion on the public health risk posed by *Listeria monocytogenes* in frozen fruit and vegetables including herbs, blanched during processing’, published in April 2020 (see Annex).

The NRL was invited to a meeting organised by the EURL to develop and validate a multi-locus sequence typing (MLST) method to potentially replace molecular serotyping; the NRL sent their apologies, as the UK uses other WGS-derived typing schemes to characterise and identify *Listeria* clusters.

FSA enquiry concerning a laboratory using an alternative method to the ISO 11290-1 for *Listeria* detection; the NRL requested further details of the alternative test and have given recommendations: investigations are still ongoing.

Coagulase-positive staphylococci

The EURL sent a call to the NRLs in January 2020 for scientific papers to be submitted for a special edition of the journal 'Toxins' entitled 'Bacterial toxins and foodborne disease', as a EURL colleague was the guest editor; the NRL circulated to relevant colleagues

An OCL required advice on the coagulase tube test to confirm staphylococcal isolates; NRL advised.

Campylobacter

In November 2019, the scientific journal 'Pathogens' requested papers for submission, as former EURL experts were guest editors for a special edition named '*Campylobacter* infections'; the NRL circulated to relevant colleagues.

An OCL requested advice regarding what other genera could be a contaminant when isolating *Campylobacter*; the NRL responded timely with expert advice.

Salmonella

In March 2020, the EURL launched an EU monitoring of S. Mikawasima isolates in food, animals, animal feed and the environment for 2020, to investigate the source of human cases. Due to the COVID-19 crisis, there will be a delay in the UK reporting data, and this will continue into the next reporting period for the NRL annual report.

Antimicrobial resistance

In December 2019, the EURL enquired of methods used by the NRLs to detect methicillin-resistant *Staphylococcus aureus* (MRSA) in food and quantify ESBL from caecal material. The NRL collated information and responded back to the EURL (the NRL do not perform MRSA in food as a routine test), whilst also copying in to the UK NRL for animal and animal feed, APHA.

The UK NRL for Food Microbiology was invited to participate in the WHO/DTU Global Sewage Surveillance Project in November 2019 for the 2020 survey but declined as the NRL knew of other organisations participating.

A survey from a Canadian institute, gathering information about initiatives and interventions to address antimicrobial resistance, as these may not be gathered in a literature review; the NRL could not complete survey, as this is not within remit or expertise

A EURL request to NRLs for methodology to detect MRSA from food samples and ESBL quantification in caecum and food samples; the NRL collated the information and responded

Related to Core Function(s): 2.a, 2.d

Representation at relevant EURL meetings and prepare meeting reports

Within this reporting period, the UK had left the EU and there have been restrictions introduced concerning attendance to EU meetings. However, PHE has been able to attend all 6 EURL meetings, either in a UK NRL capacity, or as individual experts; *Listeria monocytogenes*, coagulase-positive staphylococci (CPS), *Escherichia coli* (incl. STEC), *Campylobacter*, *Salmonella* and antimicrobial resistance (see Table 2). Where available, agendas for the meetings were forwarded to the FSA as they were received (see Annex); presentations from the UK were made at the *Listeria*, *Campylobacter* and *E.coli* meetings. Individual meeting reports were submitted to FSA after attending the meeting (see Annex).

Table 2. List of EURL meetings, January 2019 to March 2020

EURL meeting	Date: from	Date: to	Location	EURL funded	Other attendees
Listeria monocytogenes	10 April 2019	12 April 2019	Paris, France		Shona Neal
Antimicrobial resistance	25 April 2019	26 April 2019	Kgs Lyngby, Denmark		Martin Day
Salmonella	28 May 2019	29 May 2019	Amersfoort, Netherlands		Marie Chattaway
Coagulase-positive staphylococci	26 June 2019	28 June 2019	Paris, France	Amisha Vibhakar	Bruno Pichon
Campylobacter	7 October 2019	8 October 2019	Uppsala, Sweden		Frieda Jorgensen (as an individual expert)
E. coli	4 November 2019	15 November 2019	Rome, Italy		Frieda Jorgensen (as an individual expert)

Related to Core Function(s): 1.a, 2.b

Attend training workshops at the EURL

The EURLs offer a wide variety of training courses every year, and this year included detection of staphylococcal enterotoxin genes by multiplex real-time PCR and various WGS training. All EU Member States (MSs) and associated countries can show interest and attendance is decided on experience and need of the MS. However, the UK does not apply to every course, as they may have previously attended the specific training or may not perform the specific technique (for example detection of staphylococcal enterotoxin in food).

The STEC EURL invited applications to attend practical training for aspects of STEC at the EURL in Rome, Italy. The UK NRL submitted 2 applications and were both successful; the ‘Detection of STEC in food matrices according to IS/TS 13136:2012 standard and characterisation of STEC strains isolated’ held in May 2019 and the ‘Bioinformatic tools to aid STEC outbreak investigation using WGS data’ training in July 2019. Both UK participants gained personal knowledge and hands-on training; one said that it was invaluable experience for isolating STEC and the other said it was a good introduction to using the Galaxy bioinformatic tool (see Annex). Both delegates recommend the training to other colleagues.

In June 2019, MSs were invited by the AR EURL to attend a course on ‘Enhancing WGS capacity for AMR Surveillance in the EURL-AR Network’. The UK successfully secured a place and participated in September 2019. Although the participant experienced problems downloading software to the laptop, they would strongly recommend the course to other staff, and identified local improved procedures which were implemented (see Annex).

Related to Core Function(s): 2.c, 2.e

Keep abreast of methodology developments

Although the testing to detect Staphylococcus enterotoxin from food is cited in the EU microbiological criteria Regulation, 2073/2005, the UK has not had the capacity or requests to perform this on dairy products for at least 2 years. The NRL receives <1 request per year since 2013, and this makes it difficult to retain staff competency for the method and retaining accreditation. Therefore, the UK NRL outsources this method to an NRL in the Netherlands when a request is received from an OCL and it is related to Official Control work.

In April 2019, the Gastrointestinal Bacteria Reference Unit decided to discontinue the method on all other food and drink matrices due to the above reasons. This was communicated to all their customers, which included the UK OCLs effective from 1 June 2019.

Related to Core Function(s): 2.e, 2.f

Participation in the BSI AW9 microbiology committee and other working groups

In this reporting year, the BSI AW9 committee met up once in person in August 2019, and then a spill over virtual meeting in October 2019. A representative from the UK NRL for food microbiology attended the first meeting and the status of most of the related ISO standards were reviewed. The UK NRL representative was asked to contact a food microbiology laboratory in Scotland to nominate a representative on this committee, and to consider joining the WG3 Method Validation working group to provide hands-on input.

The NRL representative receives draft and final draft ISOs (DIS and FDIS) from the BSI AW9 portal throughout the year. Consequently, the NRL submitted comments for the following DIS or FDIS Standards:

- guidance on transition for the implementation of ISO 16140-3 on verification of methods
- parts 4 and 5 of the ISO 16140 series (Method validation)
- ISO 19036 (Uncertainty of Measurement)
- ISO 6579-1:2017/DAmd 1 (amendment to detection of *Salmonella*)
- ISO 7218 (General requirements and guidance for microbiological examination)
- amendments for parts 1 and 2 of ISO 10272 (*Campylobacter* detection and enumeration)
- ISO 20976-2 (Challenge tests to study inactivation potential and kinetic parameters)
- ISO 20836 (Performance of thermal cyclers)
- ISO 6887-5 (Specific rules for the preparation of milk and milk products)

A representative of the UK NRL is a member of the CEN TAG18 expert working group for the revision of the ISO TS 13136 standard (PCR detection of shiga toxin-producing *Escherichia coli*) which met in November 2019. Revision of the 2 parts of the ISO standard were discussed, which included:

- detection of *eae* may become optional in Part 1; it is important to pursue isolation of STEC for all *stx* positive samples, even when *eae* is absent
- an informative part describing dilution and/or acid treatment of enrichment before streaking/plating to enhance the growth of single colonies
- to incorporate the identification of all *stx* subtypes in Part 2

The UK NRL has nominated a representative from Campden BRI, who has expertise in challenge and shelf life studies to attend working groups organised by the *Listeria* EURL. Since the publication of ISO 20976-1 standard (Challenge tests to study growth potential, lag time and maximum growth rate) in March 2013, the *Listeria* EURL organised a working group to revise their technical guidance, to align and complement the ISO. A meeting was held in January 2020, and much progress was made.

The working group for the use of new technologies for rapid characterisation of CPS was organised by the CPS EURL. A representative of the UK NRL joined a teleconference meeting in June 2019 to discuss differences in the methods employed for detecting SE toxin genes by PCR amongst member states and then agree to develop guidance to produce harmonised approaches. However, there was limited progress in agreeing on a single platform or method to harmonise this work.
Related to Core Function(s): 2.e, 2.f, 2.g

Core Function Three: Production of standard operating procedures, codes of practice and guidance documents

Update and expand food methods archive on NRL website

At the time of writing, 11 Standard Methods and/or guidance are available on the NRL website (Table 3). These methods are based on PHE in-house methods and ISO standards, and assist OCLs to comply with the requirements of the EU Microbiological Criteria Regulations. Most of the relevant ISOs been revised under Mandate M/381, and the corresponding NRL methods are undergoing review and updates. Other relevant PHE Standard Methods have been identified that complement the NRL activities; these are also under the re-formatting process and will be archived on the NRL website. In addition, PHE SOPs are available to OCLs upon request.

Table 3. List of Standard Methods archived on the NRL website, March 2020

Document no.	Title	Version no.
FNES63	Determination of pH in food and water samples	2
FNES67	Determination of water activity in food	2
FNES8 [F12]	Enumeration of coagulase positive staphylococci (*Staphylococcus aureus* and other species)	4
FNES26 [F2]	Preparation of samples and dilutions, plating and sub-culture	4
FNES3 [F8]	Enumeration of β-glucuronidase positive *Escherichia coli*: Pour plate method	3
FNES22 [F19]	Detection and enumeration of *Listeria monocytogenes* and other *Listeria* species	4

FNES28 [F22]	Enumeration of β-glucuronidase positive *Escherichia coli* – most probable number technique	3
FNES16 [F13]	Detection of *Salmonella* species	4
FNES15 [F21]	Detection and enumeration of *Campylobacter* species	4
FNES4 [E1]	Detection and enumeration of bacteria in swabs and other environmental samples	4
FNES18 [Q4]	Guidance on Public Health response: involvement of PHE Food Water and Environmental Microbiology laboratory staff in the investigation of outbreaks of food or waterborne disease	3

Related to Core Function(s): 1.a, 1.e, 3.a, 4.a

Prepare specific guidance protocols for OCLs and the FSA

The FSA has asked the NRL to produce draft guidance for validating alternative methods in place of the reference method for the testing of food-borne organisms in food, feed and environmental samples. The ISO/CEN revision of the relevant Standard (ISO 16140) has been significant delayed, and only 3 of the 6 parts have been published at the time of writing. In addition, it is the third and fourth part that mostly influences the guidance for FSA (Part 3: Protocol for the verification of reference and validated alternative methods implemented in a single laboratory and Part 4: Protocol for single-laboratory (in-house) method validation). Pending the publication of the international standard, the NRL will finalise the guidance in 2020 to 2021.

A poor performance protocol is required if any OCLs generate repeated poor results from the European Food Microbiology Legislation (EFL) External Quality Assessment Scheme. However, there has been no consistent poor performance from the participant results (see next section).

Related to Core Function: 2.a, 2.d, 3.a, 4.a

Manuscript of 4 years of OCL performance of the EFL scheme

There has been some progress made but it is not yet completed, due to other work priorities. Therefore, this activity is transferred to the 2020 to 2021 work programme.

Related to Core Function: 3.a, 4.b

Core Function Four: Compliance assessment via audits and ring trials

OCL participation in the European Food Microbiology Legislation Proficiency Testing Scheme

The NRL is required by Regulation (EC) 625/2017 to organise and assess performance of OCLs through relevant comparative testing such as interlaboratory studies.

In February 2019 the UK OCLs performing microbiological tests on food were invited to register to the 2019 to 2020 European Food Microbiology Legislation (EFL) External Quality Assessment Scheme, as provided by the PHE Food Environmental Proficiency Testing Unit (FEPTU). Based on the requirements of EU Regulation 2073/2005 (as amended) for the microbiological criteria for foodstuffs, the scheme enables the performance assessment on the identification, examination and interpretation of microbiological results of samples tested against these legislative criteria. A full scheme comprises of 12 samples from 4 distributions based on food categories within the regulation and results are submitted on a web-based form. Further details can be found at this link: https://www.gov.uk/government/collections/external-quality-assessment-eqa-and-proficiency-testing-pt-for-food-water-and-environmental-microbiology#european-food-microbiology-legislation-scheme.

Samples processed from the EFL Scheme continue to be a test for participating laboratories in maintaining their capability with knowledge of microbiological-criteria and changes made to legislation that is driven by evidence-based scientific opinion and emerging risks in foodborne disease. The scheme not only provides a single suitable platform to assess microbiological test proficiency, but also enables the demonstration of laboratory understanding when it comes to showing compliance against legislative requirements, as laboratories who test a variety of samples for official control should be able to apply. Decisions on testing should be based on fact, not assumption and should refer to the FEPTU guide to scoring for more information.

The NRL supports OCL participation of this scheme which allows direct performance comparisons across the OCLs but also acts independently from the scheme organisers. Results are anonymised, and reports do not disclose the identity of any laboratory. The NRL monitors the performance of each laboratory and invite laboratories to seek assistance from the NRL when experiencing difficulties.

Thirteen out of 14 OCLs registered to participate to the full scheme in 2019 to 2020 and overall results were satisfactory. Proficiency for achieving the correct microbiological

results continue to be satisfactory for the majority of testing; the final distribution (EFL 50, dairy foods) did see a spike of incorrect results and generated the highest number of wrong results for the whole year of 5 (3 low enumerations and 2 false positive results). The 5 incorrect results were reported by different laboratories and no trend was observed for any sample or test. Not all laboratories examined samples in every distribution received; the NRL requests laboratories to only sign up to those matrices that are relevant to the laboratory testing capabilities or if it would enable laboratory training.

Table 5 summarises the samples for 2019 to 2020 and the performance of OCLs that carried out the examinations.

This 2019 to 2020 distribution included the challenge of tests and methods that OCLs have either recently implemented or have no capacity. These were staphylococcal enterotoxin, STEC detection and enumeration of *Campylobacter* and improvements in specific areas for these procedures were observed.

The 'referral of tests' option should be applied for examinations that would not normally be performed in the OCL, but this was seldom used by laboratories this year, despite opportunities with samples where this option would have been appropriate and correct. Staphylococcal enterotoxin testing was required on sample EFL146 (unpasteurised soft cheese at manufacture) since levels of coagulase-positive staphylococci were greater than 10^5 cfu/g in the sample. Whilst 4 laboratories correctly identified the need for this test, no laboratories stated that they would refer this examination and therefore not demonstrating their normal laboratory procedures.

Table 5. Overview of performance of the 2019 to 2020 European Food Microbiology Legislation Scheme

Sample code	Brief sample details	Required examination(s)	OCLs achieving >70% of the maximum possible score of 8[1]
EFL145	Ready-to-eat cottage pie meal made from beef meat, sampled before the product has left the immediate control of the manufacture	*L. monocytogenes* detection	12/12
EFL146	Soft fresh cheese made from unpasteurised milk, sampled during manufacturing process	*L. monocytogenes* detection Coagulase-positive staphylococci Staphylococcal enterotoxins detection	12/12 11/11 0/0[2]
EFL147	Frozen ice-cream made from pasteurised milk. Product placed on the market during shelf-life	*L. monocytogenes* enumeration	11/11

EFL148	Pre-made liquid pancake mix made from pasteurised milk and egg, sampled at the end of manufacturing process	*Enterobacteriaceae*	10/11
EFL149	Ready-to-eat spring rolls made from rice paper sheets containing primarily raw alfalfa sprouts - shelf-life <3 days. Placed on market during shelf life	*L. monocytogenes* enumeration *Salmonella* spp. Shiga-toxin *Escherichia coli* detection	11/11[3] 11/11 4/4[4]
EFL150	Ready-to-eat pre-cut watermelon with a shelf life of >5 days, taken during the manufacture process	*L. monocytogenes* detection *Escherichia coli*	10/11 10/11
EFL151	Cold smoked speck ham with herb seasoning. Product placed on market during shelf-life	*L. monocytogenes* enumeration *Salmonella* spp.	12/12 10/10
EFL152	Raw minced lamb to be eaten cooked. Placed on the market at the end of manufacturing process	*Salmonella spp.* Aerobic Colony Count *Escherichia coli*	7/7[5] 12/12 12/12
EFL153	Broiler carcase, product placed on the market after the chilling process	*Salmonella spp.* *Campylobacter spp.*	10/10 6/6[6]
EFL154	Dried dietary foods for special medical purposes intended for infants below 6 months of age. Product placed on the market during shelf-life	*L.monocytogenes* detection *Salmonella* spp. *Cronobacter* spp.	10/11 12/12 1/1[7]
EFL155	Liquid body building whey supplement – shelf-life is < 5 days. End of manufacturing process	*L.monocytogenes* detection *Enterobacteriaceae* Coagulase-positive staphylococci	7/8 11/11 2/3[8]
EFL156	Cottage cheese made from raw milk, sampled during the manufacturing process	*L.monocytogenes* detection Coagulase-positive staphylococci Staphylococcal enterotoxins detection[9]	12/12 9/10

[1]Number of laboratories achieving >70% compared to the total laboratories participating in the examination. Those that did not return any data or did not examine samples were not included in this table.

[2]Four laboratories correctly identified the food category.

[3]Eight laboratories incorrectly identified the food category for *Listeria* according to changes made to the regulation for sprouted seeds able to support the growth of *Listeria monocytogenes* (Amd 2019/229) but were awarded points on this occasion.

[4]Ten laboratories correctly identified the food category.

[5]NRL are investigating low performance for this test.

[6]Eight laboratories correctly identified the food category for *Campylobacter* spp. for this sample.

[7]Nine laboratories correctly identified the food category for *Cronobacter* spp. for this sample.
[8]More than 1 food category was applicable to this sample; CPS test is required for category 2.2.7
[9]Selection of test dependant on level of coagulase-positive staphylococci reported; marks for staphylococci enterotoxin were not awarded

The same 'referral of tests' omission was observed for STEC. Not only was there a slight decline in the number of laboratories able to perform STEC testing; 4 laboratories reported a result for sample EFL149 (ready-to-eat spring rolls containing raw sprouts) compared to 5 laboratories performing this test in June 2017 (EFL121, ready-to-eat salad containing bean shoots); no laboratories chose to refer the test to another laboratory. However, most laboratories did continue to demonstrate their compliance for this criterion, with 10 laboratories again stating the right food category from the food safety criteria.

Test referral was not completely absent; *Cronobacter* spp. detection was required for sample EFL154 and 9 laboratories correctly identified this test. However, only 2 laboratories reported referral for identification of colonies. Laboratories are reminded that the option to refer remains in the scheme design and should be used for tests and samples that cannot be performed in-house and that would normally be sent away.

EFL149 also saw a failure of laboratories correctly classifying this sample for *Listeria*. An amendment to the legislation in 2019 (EU 2019/229) was issued that reclassified sprouted seeds as a food able to support the growth of *Listeria monocytogenes* and changes made to footnote 4 in the annexe. Eight laboratories incorrectly stated category 1.3 (3 stated 1.2) and therefore incorrectly classified this sample as would not support the growth of this organism. Laboratories were awarded points on this occasion for the incorrect food category due to an error in the reporting of the sample but were reminded to read the annex of amendments especially where deletions or changes may not be evident.

The EU microbiological criteria are not clear on samples taken at the end of manufacture and with a shelf life of less than 5 days. EFL155 (liquid body building whey supplement at end of manufacture) was therefore difficult to place for *Listeria* requirements; 6 laboratories decided that this would support growth of *Listeria monocytogenes* despite a short shelf-life and 5 laboratories did not identify *Listeria* at all.

An improvement was seen in laboratories able to enumerate for *Campylobacter*, as observed in sample EFL153 (broiler carcase post-chill). Although 8 laboratories identified the food category for this sample, 6 laboratories did perform examination and reported a correct result. This is compared to sample EFL139 in January 2019, where 10 laboratories identified *Campylobacter* enumeration and 5 laboratories performed satisfactory testing.

Sample EFL152 saw few laboratories correctly identifying *Salmonella* spp. This was a sample of raw minced lamb to be eaten cooked and sampled at the end of manufacture. Only 7 laboratories identified criterion 1.6 and correctly report the result, thus scoring the maximum marks available. Five laboratories however, omitted the *Salmonella* test, but did test for the other correct parameters for this sample. The NRL are in discussion with FEPTU to understand the reasons for this unusually low performance and will inform OCLs of the outcome.

The NRL will continue to assess laboratory performance through proficiency testing and has invited all OCLs to register to the EFL scheme for the 2020 to 2021 distributions (see Annex). From December 2019, OCLs are obliged to participate, on request by the NRL, and should aim to comply with the new regulation in force (EU Reg. 625/2017). Participation of OCLs will provide overall assurance of laboratory competence, identify areas of weakness and further training as well as support compliance with laboratory quality standards and accreditation. OCLs will also continue to have access to expert advice and support from FEPTU and/or the NRL.

Related to Core Function(s): 4.a, 4.b

Participate as UK-NRL in EURL ring trials and other initiatives (2019 to 2020)

The NRL is mandated to collaborate with and participate in inter-laboratory comparative tests as organised by the European Reference Laboratories in each of the areas that the NRL are responsible for. In 2019 to 2020, the NRL has received 10 ring trial distributions from 5 EURLs, that covers the work carried out by OCLs and the reference laboratories in the UK, and includes tests for detection, enumeration and strain typing. Table 6 lists these activities and a summary of performance.

The EURL is increasingly providing the option in their trials to perform typing methods by Whole Genome Sequencing (WGS) as this becomes the method of choice for Member States. Typing trials for *Campylobacter*, *Listeria monocytogenes*, Shiga-toxin producing *Escherichia coli* and *Salmonella* were all performed by the NRL using WGS and satisfactory results were produced. After receiving a panel of pure culture, DNA material or sequencing files from the EURLs, assessment of proficiency included the correct subtyping of strains, detection and typing of virulence genes and serogroups and the cluster analyses of strains. WGS was introduced for routine reporting of gastrointestinal bacteria typing results in the UK since 2015, starting with *Salmonella*, and is now employed routinely for all organisms that the NRL has responsibility for official control.

The NRL did not perform typing and characterisation for antimicrobial resistance in *Salmonella*, *Campylobacter*, staphylococci, enterococci or *Escherichia coli* this year, since the UK NRL for food microbiology adopts a different method to that stipulated in

EU legislation (a broth dilution method whereas the UK NRL performs an agar dilution method). APHA are the UK animal and feed NRL for these activities and perform the sampling and analysis activities as laid down in Decision EU 652/2013 for the monitoring of antimicrobial resistance. As the official laboratory network in the UK for this activity, APHA participated in the trials for antimicrobial resistance. Results from APHA are reviewed at regular liaison meetings and lessons learnt from all EURL PTs are communicated to OCLs by email or though the NRL quarterly newsletter.

Proficiency tests for the enumeration and detection of organisms in food and environmental matrices were organised by 5 of the EURLs and the UK NRL scored satisfactorily in tests for the enumeration and detection of *Campylobacter* in chicken meat, detection of *Salmonella* in flaxseeds and detection and isolation of STEC from flour (Table 6). Although the NRL was excluded from the performance analyses for the enumeration of coagulase-positive staphylococci for deviating from the reference method (ISO 6888-1 standard sets counting limits on plates with typical growth), the results achieved were comparable to those intended for all the samples. The UK NRL results for enumerating *Listeria monocytogenes* in environmental swab samples were also not analysed by the EURL for performance due to a discrepant result in the low-level sample and which the NRL is investigating as a non-conformance.

The EURL for *E.coli* mandated their PT for the detection and isolation of STEC to be performed using only buffered peptone water (BPW). The reference method, ISO/TS 13136, prescribes the use of modified soy broth (mTSB) for samples with a high background of organisms, but has been observed to hamper the growth of certain STEC strains. The use of BPW would be more appropriate and validation is on-going for collating data through ring trials and proficiency programmes and comparing results between the 2 broths. The UK NRL participated, and correctly reported the virulence gene assets of the serogroup and isolated the STEC strain from all samples of flour that were contaminated (Table 6). Results of these trials will be used to inform the revision and improvement of the reference method which is also mandated to become a full international standard.

The test for detection of staphylococcal enterotoxins in milk and cheese samples was declined by the UK NRL this year. Demand for toxin detection in coagulase-positive staphylococci in the UK is very low, with on average 1 request for testing made every 2 years. Sustaining the capability and competency in the methodology for this infrequent examination is difficult and with agreement from the FSA, a justification by the UK NRL for not participating in the proficiency trial was submitted and accepted by the EURL for CPS. The NRL does maintain a testing facility for samples by sub-contracting the service to a laboratory who themselves are a designated official control laboratory within their country and the NRL network and participate in the EURL proficiency testing. OCLs have been communicated on procedures for requests for this testing through the June 2019 newsletter and the last NRL User day and should contact the NRL for advice and further information.

Related to Core Function(s): 1.a, 2.e, 4.c, 4.d

Table 6. NRL participation in EURL ring trials, March 2019 to February 2020

Month received	Organism – test[1]	Reference	Matrix/pure culture	Comments
March 2019	*Campylobacter* – enumeration and voluntary species identification in food	PT23	Chicken meat	Satisfactory performance for enumeration (100%); lower performance for species identification (62.5%) due to limitations in methodology
March 2019	*Campylobacter* – detection and species identification	PT24	Chicken meat	Satisfactory performance for detection (100%); lower performance for species identification (75%) due to limitations in methodology
March 2019	*Campylobacter* – typing	PT25	DNA	Satisfactory performance (100%)
March 2019	*Listeria monocytogenes* – enumeration	Anses_LSAl_19_01_Listeria	Swabs	Excluded from performance due to discrepant result: results indicate satisfactory performance for high level sample and unsatisfactory performance for low level sample
March 2019	*Salmonella* – detection in food	5th	Flax seeds	Satisfactory performance (100%)
June 2019	*Listeria monocytogenes* – typing	7th	Pure cultures	Excluded from performance due to not performing all prescribed methods of the PT. WGS results indicate 100% match with the intended results
October 2019	CPS – enumeration in food	Anses_LSAl_19_08_LRUE_CPS	Mozzarella cheese	Excluded from performance due to deviations from the reference method but results indicate satisfactory performance compared to the intended result
October 2019	STEC – detection and isolation in food	PT25	Flour	Satisfactory performance
October 2019	STEC – typing	PT26	Pure cultures	Satisfactory performance
November 2019	*Salmonella* – serotyping	Serotyping 2019	Pure cultures	Satisfactory performance (100%)

[1]STEC = Shiga toxin-producing E. coli, CPS = Coagulase-positive staphylococci

Organise a Skype-based workshop on Implementation of ISO 17025:2017 for UK OCLs

The NRL organised a training session using Skype in April 2019 concerning the implementation of the revised ISO 17025:2017 (see Annex for agenda). The UK Accreditation Service requires that all testing laboratories should transfer their accreditation to the new version of the Standard, and the 3 speakers who presented from the PHE FW&E Network included an introduction and best practices for a good quality system, the Standard's main changes from the previous version, and how 2 of the PHE laboratories experienced the assessment and gained accreditation. Overall, the session went well, with 23 colleagues participating from 7 OCLs and the FSA. Whilst there were problems with the technology, feedback was very good, and participants felt the session was open and relevant to their needs.
Related to Core Function(s): 2.a, 3.a, 4.e

Organisation of a practical workshop for UK OCLs – Skype session for Uncertainty of Measurement and Principles of PCR

Feedback from previous NRL events indicated that practical workshops scored less than other theoretical learning. Therefore, the NRL organised a second Skype training session on Uncertainty of Measurement (UoM) in November 2019. The national method and calculation spreadsheet was published earlier this year and these 2 documents were demonstrated, along with the newly revised ISO 19036:2019. Over 30 people attended via Skype and there were useful discussions, including the use of the spreadsheet for non-microbiological data (e.g. water activity) and the absence of legal guidance for UoM acceptability limits. With good feedback from the participants, it was noted that a follow-up session on UoM should be arranged in the next 2 years.

A further Skype session on 'Principles of PCR and Real-time PCR in Food Microbiology' was held in January 2020, as feedback from other workshops indicated that learning about the fundamentals of PCR was a useful addition to expand their knowledge. Twenty-two people attended an hour-long presentation, which included chemistry of DNA extraction and PCR, comparing block-based and real-time methods, preventing cross-contamination, interpreting results and applying them to the microbiological criteria, (EC) 2073/2005. Future topics of training that the NRL could deliver to expand shared knowledge and fulfil any training gaps will be based on OCL feedback.
Related to Core Function(s): 2.a, 3.a, 4.e

Core Function Five: Co-ordination within the UK of EURL initiatives

Support food aspect of the EU-wide AR monitoring (Decision 2013/652/EU)

Fresh meat at retail have been sampled and tested for the above EU Decision in the UK by the APHA, since 1 January 2015. The NRL have been available for support and advice to the APHA and FSA. There has been continuation by APHA in sampling and testing the retail component of the EU harmonised survey for the FSA in the reporting year.

In addition, the revision of EU Decision 2013/652/EU is still in progress and is due to be in force by 1 January 2021. This has generated questions from the EURL for methodology in December 2019, which was detailed in Core Function Two; Provide impartial advice to FSA, OCLs and other UK laboratories, and required liaising with APHA.

Additional information regarding APHA liaison and AR work can be found above in Core Function One; Liaise with APHA regarding mutual NRL activities.

Related to Core Function(s): 1.b, 2.a, 5.a

Participate in EURL activities relating to Whole Genome Sequencing

A mandate was sent from the EC to EFSA and ECDC in 2017 to expand the molecular typing data collection to WGS data. A working group was then formed by EFSA, ECDC and the EURLs focused on collating WGS capacity from the Member States, to assess the bioinformatic pipelines available, the requirements necessary to analyse WGS data, and the support needed to implement WGS into MSs (for example, training, methods). This was followed up by a letter from the EU requesting EFSA and ECDC to implement the collection and analysis of WGS by June 2022.

As part of this work, a conference ‘Modern technologies to enable response to crises: Next Generation Sequencing to tackle food-borne diseases in the EU’ was organised by the *E.coli* EURL. This was due to be held on 10 March but has been postponed to 25 September due to COVID-19.

Annex – Documents produced from NRL activities

Core Function One: Secretariat services	
Dissemination of information from the EURLs *Related to Core Functions: 1.a, 1.c, 2.d, 2.e, 2.f, 4.c, 5.a*	• EURL Lm_2019_2020_WP • WP_EURLCPS_2019 to 2020_V1.2 • 2019-20 Salmonella EIURL Work Programme_8 Kirsten Closure 190529 • EURL-Salmonella Newsletter June 2019_0 • EURL-Salmonella Newsletter September 2019 • EURL-Salmonella Newsletter December 2019 • EURL-Salmonella Newsletter March 2020 • 524_2019-dec-newsletter-no13-final
Quarterly newsletters *Related to Core Functions;* *1.a, 2.d, 2.e, 2.f*	• NRL_newsletter_Q1_2019 • NRL newsletter Q2 2019 • NRL newsletter Q3 2019_FINAL-1 (3) • NRL newsletter Q4 Mar20_final
Coordination of 2018 OCL User Day *Related to Core Functions:* *1.a, 1.b, 1.c, 2.a, 2.d, 2.e,* *2.f*	• NRL_newsletter_Q1_2019 • NRL newsletter Q2 2019 • NRL newsletter Q3 2019_FINAL-1 (3) • NRL newsletter Q4 Mar20_final
Coordination of 2018 OCL User Day *Related to Core Functions: 1.a, 1.b, 1.c, 2.a, 2.d, 2.e, 2.f*	• User day 2019 agenda_18JunSN,KL (FINAL)
2019 Audit – OCL's capabilities and requirements *Related to Core Functions: 1.b, 2.d, 2.e, 4.a*	• Select Survey Questions Sent to all OCLs re 2019 OCL Audit
Provide regular updates to FSA *Related to Core Functions: 1.d*	• FSA Meeting Mins 24Jun19 (Final)_ KL_SN2_Avedits • FSA Meeting Mins 17Sept19_KL_SN_AV_JMcL_edits, MS (FINAL) • FSA Meeting Mins 04Dec19_KL_SN_AV_JMcL_edits (FINAL) (002) MG_VM comments • FSA NRL Meeting 30Mar20 (Final Draft) KL_SN_Mgedits • UK NRL Monthly Log_APR2019_KL_AV_SNedits (FINAL) • UK NRL Monthly Log_MAY2019_FINAL • UK NRL Monthly Log_JUN2019_Final • UK NRL Monthly Log_JULY2019_FINAL_AV_KL_SN_AV_JMcL edits • UK NRL Monthly Log_AUG2019_Final • UK NRL Monthly Log_SEPT2019_final • UK NRL Monthly Log_OCT2019_final • UK NRL Monthly Log_NOV2019_Final • UK NRL Monthly Log_DEC2019_Final • UK NRL Monthly Log_JAN20_FINAL_AV_Sna • UK NRL Monthly Log_FEB20_FINAL • UK NRL Monthly Log_MAR20_Final

Core Function Two: Advice and representation within the UK/E	
Provide impartial advice to FSA, OCLs and other UK laboratories *Related to Core Functions: 2.a, 2.d*	• j.efsa.2020.6092_Lm in frozen fruit and veg
Representation at relevant EURL meetings and prepare meeting reports *Related to Core Functions: 1.a, 2.b*	• AgendaLm2019 (003) • Internal report of EURL meeting Lm 10-12 April_final • EURL Lm_report WS Lm 2019 • 525_eurl-ar-ws-2019-minutes-final • Programme of workshop 2019 v.190424 • Indvivdual Report on the 24th Salmonella EURL Workshop 2019 • 2019-0135 • AgendaCPS2019_2 • Internal report of EURL meeting - CPS Workshop June 2019 • EURL CPS_report 2019 workshop • Workshop program 2019 • Report_EURL-WS 2019 • Agenda_WS
Attend training workshops at the EURL *Related to Core Functions: 2.c, 2.e*	• Evaluation of EURL training_ISO 13136 KA May 2019 • Evaluation of EURL Training_WGS Outbreak investigations_AI • MG_AR Training 24_29 Sept19_Evaluation
Core Function Three: Production of standard operating procedures, codes of practice and guidance documents	
Update and expand food methods archive on NRL website *Related to Core Functions: 1.a, 1.e, 3.a, 4.a*	https://www.gov.uk/government/collections/uk-national-reference-laboratory-for-food-microbiology
Core Function Four: Compliance assessment via audits and ring trials	
OCL participation in the European Food Microbiology Legislation Proficiency Testing Scheme *Related to Core Functions: 4.a, 4.b*	• FEPTU483.11_Schedules and prices 2020 to 2021
Organise a Skype-based workshop on implementation of ISO 17025:2017 *Related to Core Function: 2.a, 3.a, 4.e*	• ISO 17025 programme 25Apr19 - draft (004)
Organise a Skype session for UoM and Principles of PCR *Related to Core Function: 2.a, 3.a, 4.e*	• UoM Programme - 19_ final • PCR principles

Proposed PHE NRL Activities, April 2020 to March 2021

1 Core Function One: Secretariat services

1.a. Disseminate information/advice from international organisations to FSA, OCLs and other UK laboratories in a timely and effective manner.

1.a. Produce and circulate quarterly newsletters to FSA, OCLs and other UK laboratories.

1.a. Co-ordinate the OCL User Day to update UK OCLs and other relevant UK laboratories of the NRL core functions.

1.a. Assist in the dissemination and advice of EPIS and other alerts from the appropriate authority.

1.b. Analyse and publish the audit results regarding the OCL's capabilities and requirements

1.b. Review content of the UK Food Examiner Register.

1.b. Continue liaison meetings with APHA for AR, *Campylobacter* and *Salmonella*.

1.c. Liaise with FSA in matters arising from implementing the new Official Control Regulation, (EU) 2017/625

1.c. Liaise with Campden BRI to discuss and implement challenge testing activities

1.c. Liaise with CEFAS for any overlapping NRL activities

1.d. Provide regular updates to the FSA on NRL activities by producing monthly reports and meet on a quarterly basis.

1.d. Produce and submit annual report to the FSA on NRL activities for 2020 – 2021.

1.e. Maintain and update the NRL web content on the PHE website.

2 Core Function Two: Advice and representation within the UK and internationally

2.a. Provide impartial expert advice to FSA, OCLs and other UK laboratories, upon request.

2.b. Represent the UK at relevant international meetings and working groups; consult FSA prior to meetings and submit an internal report after attendance of meetings.

2.c. Attend training workshops at international organisations, where relevant and after successful applications.

2.e. Keep abreast of methodology developments and advise FSA and OCLs (for example, Service Level Agreement for CPS toxin testing).

2.g. Participate in the BSI AW9 microbiology committee.

2.g. Participate in Working Group to revise the ISO/TS 13136:2012 (PCR detection of STEC).

3 Core Function Three: Production of standard operating procedures, codes of practice and guidance documents

3.a. Update and expand food methods archive on NRL website.

3.a. Prepare a guidance document for OCLs and the FSA on the use and validation of alternative methods for testing Official Controls.

3.a. Produce a poor performance protocol for OCL participation in the EFL proficiency test scheme.

3.a Draft a manuscript for peer-review summarising 4 years of OCL performance of the EFL PT scheme.

4 Core Function Four: Compliance assessment via audits and ring trials

4.a. Ensure consistency and quality of testing approached applied by UK OCLs and support where necessary.

4.b. Liaise with FEPTU and monitor OCL’s comparative testing performance and assist OCLs in the implementation of corrective measures.

4.b. Evaluate OCL’s performance using trend analysis.

4.c. Coordinate the participation of OCLs in international method validation studies and other initiatives and report to FSA.

4.d. Participate as UK-NRL in proficiency tests and method validation studies organised by the EURL (where available) and report to FSA

4.e. Organise Skype/classroom-based workshops for UK OCLs, dependent on the outcome of the audit (1.b) and OCL needs.

4.e. Organise a practical workshop for UK OCLs, dependent on the outcome of the audit (1.b) and OCL needs.

5 Core Function Five: Coordination within the UK of international initiatives

5.a. Support the food aspect of the EU-wide AR monitoring (Decision 2013/652/EU), liaising with FSA, OCLs, relevant Reference Laboratories and APHA.

5.a Liaise with APHA, audit and review strategy for harmonization of existing antimicrobial resistance testing.

5.a Provide information regarding Whole Genome Sequencing when requested from the EURLs, and participate in any related workshops, proficiency testing, training and guidance documentation, where necessary.

Summary of NRL activities for April 2019 to March 2020

Activities	Function	Core	0	1	2	3	4	5	6	7	8	9	10	11	12
			March 2019	April 2019	May	June	July	August	Septembe	October	Novembe	Decembe	January 2020	February	March
Produce & circulate quarterly newsletter to FSA, OCLs & other labs	Disseminate information	1.a													
OCL User Day meeting at NRL, 18th	Meeting	1.a													
Perform OCL Survey for capabilities and capacity	Secretariat	1.b													
Review content of UK Food Examiner register	Secretariat	1.b													
Continue liaison meetings with APHA Salmonella, Campylobacter and AMR NRLs	Secretariat	1.b													
Liaising with FSA matters implementing (EU)2017/625	Advice/communication	1.c													
Annual report to FSA	Coordination	1.d													
Meetings with FSA	Coordination	1.d													
Monthly reporting to FSA	Coordination	1.d													
Maintain and update NRL web content on PHE website	Website	1.e													
Listeria 13th Workshop in Anses, Paris, 10-12th	EURL Workshop	2.b													
Antimicro Resist 13th Workshop in Copenhagen, 25-26th	EURL Workshop	2.b													
STEC detection and typing from food training, 13-17th	VTEC EURL Training	2.c													
Salmonella Workshop in Uppsala, Sweden, 28-29st	EURL Workshop	2.b													
Coag+ Staph 13th Workshop in Anses, Paris, 26-28th	EURL Workshop	2.b													
Campy 14th Workshop in Uppsala, Sweden, 7-8th	EURL Workshop	2.b													
Organisation of Campylobacter PT training, 9th	Campy EURL Training	2.c													
E.coli 14th Workshop in Rome, 4-5th	EURL Workshop	2.b													
Enumeration, detection and species identification of Campylobacter spp training, 12-15th	Campy EURL Training	2.c													
Agree the workflow and Service Level Agreement for CPS toxin testing with the Dutch NRL	Advice	2.e													
Participation in WG for revised ISO 13136 (STEC)	Advice & representation	2.g, 5.a													
Participation in BSI AW9 microbiology committee	Advice & representation	2.g													
Produce guidelines on alternative methods	Guidance document	3.a													
Produce poor performance protocol for OCL PT participation	PT document	3.a													

Activities	Function	Core	0	1	2	3	4	5	6	7	8	9	10	11	12
			March 2019	April 2019	May	June	July	August	Septembe	October	Novembe	Decembe	January 2020	February	March
Produce OCL performance review of 4 years' participation of EFL scheme	Peer-reviewed manu	3.a, 4.b													
Revise and publish UK SOPs on website	Maintain SOPs	3.a													
Liaise with FEPTU & monitor OCL's testing of EFL scheme	UK PT	4.b													
Coordinate participation of OCLs in international method validation studies	International ring trials	4.c													
Campy enumeration in chicken meat PT from EURL (PT23)	EURL PT	4.d													
Campy detect & species id in chicken meat PT from EURL (PT24)	EURL PT	4.d													
Campylobacter subtyping PT from EURL (PT25)	EURL PT	4.d													
Listeria enumeration in environmental samples from EURL	EURL PT	4.d													
Salmonella detection in flaxseeds PT from EURL	EURL PT	4.d													
VTEC detection in sprouts from EURL (PT 21)	EURL PT	4.d													
Coag+ Staph toxin detection PT from EURL	EURL PT	4.d													
Coag+ Staph enumeration in food EQA from EURL	EURL PT	4.d													
AMR for *E. coli*, enterococci and staphylococci EQA from EURL	EURL EQA	4.d													
VTEC detection in sprouts from EURL (PT 25)	EURL PT	4.d													
AMR Salmonella & Campylobacter EQA from EURL, TBC	EURL EQA	4.d													
E.coli & VTEC vir gene typing and WGS PT from EURL (PT-PFGE8;PT26;WGS3)	EURL EQA	4.d													
Salmonella typing EQA from EURL	EURL PT	4.d													
Organise practical workshop for UK OCLs (TBC)	Workshop	4.e													
Organise Skype workshop on implementation of ISO 17025:2017 for UK OCLs	Workshop	4.e													
Support food aspect of EU-wide AR monitoring (Decision 2013/652/EU)	EU monitoring	5.a													
Provide information regarding WGS processes to EURLs and other international organisations	EURL initiative	5.a													