

15th EURL-AR Workshop, 21-22 September/2021

(virtual meeting)

Minutes

The minutes are listed according to the agenda.

Presentations may be found on the EURL-AR website (<https://www.eurl-ar.eu/presentations/workshop-virtual-meeting-september-2021.aspx>). Specific links to presentations are imbedded below to the extent that the speaker have allowed for the publication of their slides.

Participants

Invitation for participation was circulated to the EURL-AR Network, the UK NRL was invited to participate in the EQAS component in the morning of the 22 September.

From the EURL-AR-network, all EU member states (MS) were represented at the workshop except for Croatia. Participating non-MS connected to the EURL-AR network were Iceland, Norway, Serbia, Switzerland, and Turkey. For the component of the meeting related to the EURL-AR EQAS, representatives from the United Kingdom participated. Additionally, the EU Commission and EFSA were represented.

Approx. 95 participated online 21 September 2021 and approx. 85 participated online 22 September 2021.

Tuesday, 21 September 2021

Welcome and introduction (Rene Hendriksen, EURL-AR)

Update from the EURL-AR (Rene Hendriksen, EURL-AR). See presentation ([Link](#))

The EURL-AR aims to provide scientific/technical assistance for the EU Commission regarding AMR monitoring and this includes various projects and organizations (WHO, GLASS, FAO, UNEP, OHEJP, FF etc.).

Over the past year, specific EURL-AR activities included

- A survey regarding pre-enrichment procedures for ESBL, AmpC and carbapenemase producing *E. coli*
- A webinar regarding WGS procedures after which the WGS protocol was updated
- Release of a new updated version of the online tool ResFinder (v4.1) and a scientific paper published in Jul 2020, which describes the new beneficial features
- A webinar regarding the new Sensititre™ plates (EUVSEC3 and EUCAMP3) for the EU monitoring of AMR was arranged together with Thermo Fisher, Nov. 2020
- Various proficiency tests: 1) Phenotypic trial (*Salmonella*, *E. coli* and *Campylobacter*), 2) EQA Matrix (2020) (ESBL, AmpC or carbapenemase producing *E. coli*) and 3) Genomic PT 2020 (*Salmonella*, *E. coli* and *Campylobacter*)
- Webinar in which Martial Plantady presented the new AMR Decision (1729/2020/EU) and border control for member states (MS)
- Webinar related to harmonized *Campylobacter* protocol
- The EURL training course (virtual) in spring 2021 which included exercises in QC of NGS data, AMR gene detection, pheno- and genotype concordance and phylogeny. The course had 170 participants from 37 laboratories

Update from EFSA (Pierre-Alexandre Beloeil, European Food Safety Authority)

See presentation ([Link](#))

Pierre-Alexandre presented slides. Over the past year activities at EFSA of specific interest for the EURL-AR have been that EFSA has produced the 2019 EUSR on AMR, prepared the 2021 data reporting (Guidance docs), EFSA AMR ENV Scientific Opinion (B. Guerra), EFSA WGS platform for supporting AMR data collection (M. Rossi) and issued the JIACRA III report in June.

There has been an Interagency collaboration regarding relationship in human/animals between antimicrobial consumption (AMC) and AMR over a number of years (2001-2018) and the overall observations this year are that:

The overall AMC was lower in food-producing animals than in humans (27 EU/EEA countries) over the 2016-2018 period. A statistically significant decrease of 32% in the population weighted mean AMC in food-producing animals between 2014 and 2018 was observed.

Antimicrobial consumption varied by country. In 2017, among 29 EU/EEA reporting countries: 1) in 20 countries, AMC was lower, 2) in 1 country, AMC was similar and 3) in 8 countries, AMC was higher, in food-producing animals than in humans.

Aminopenicillins, 3rd- and 4th-generation cephalosporins and quinolones were used more in humans than in food-producing animals whereas polymyxin b and tetracyclines were used more in food-producing animals than in humans.

There are links between AMC in animals and AMR in bacteria from food-producing animals, which in turn is associated with AMR in bacteria from humans. For example, *Campylobacter* spp. bacteria in food-producing animals and in humans.

In most countries, the key AMC indicators decreased, both for food-producing animals and in humans.

A statistically significant negative association was observed between the primary key indicators in food-producing animals, AMC and the occurrence of completely susceptible indicator *E. coli*. A clear and consistently lower probability of detecting completely susceptible indicator *E. coli* was observed when AMC was higher.

Conclusion: Further interventions to reduce AMC will have a beneficial impact on AMR. We need to promote, in both humans and food-producing animals: 1) prudent use of antimicrobial agents, 2) infection control and 3) prevention of infection. High levels of AMC and AMR are still being reported.

Summary of the plenary discussion:

A question was posed in relation to follow-up on specific MS, whether targeted action would be taken. EFSA stated that as for mitigation actions, the recommendations are still of actuality and implementation should continue. We are aiming in the right direction and the legislations appears to be in place in order to collect even more interesting data.

Update from the European Commission and implementation of monitoring decision 2020/1729/EU (Martial Plantady, European Commission)

The new AMR Decision (1729/2020/EU) has replaced the former by 1/1-2021 and MS have applied the guidelines in the new decision for approx. 9 months. In general, this new decision is close to the former with few essential updates and so far no difficulties from any MS has been reported. AMR monitoring is in line with the former decision regarding test bacteria and food production animals. Border control sampling is new but sampling procedures are the same. The EUVSEC panels were slightly changed. Another novel implementation in the new decision is that it is now possible to apply WGS as an analysis method. In addition, it has been decided that the EU Commission will no longer be in charge of the financial aspects which is now administrated by HaDEA. A questionnaire regarding this will be forwarded to the MS.

Summary of the plenary discussion:

To have an impression of how many countries in the EURL-AR network that are currently using WGS, the workshop participants were asked to indicate in the chat if they used WGS. Six countries (DK, ES, FIN, DE, NO, NL) replied that they will provide NGS data in 2021 whereas two countries (LU, AT) will start up WGS in 2022. Moreover, Belgium will potentially start to produce WGS data regularly in the future and notably all countries aim to run traditional MIC testing in parallel with WGS.

It is now possible for the MS to have expenses for 40 EUR/isolate covered in relation to storage of isolates for 5 years.

Rationale for the changes in the antimicrobial panels (Rene Hendriksen, EURL-AR)

See presentation ([Link](#)).

The presentation intends to give an update on why the antimicrobial panels for the AMR monitoring have been changed. EFSA identified an expert group to identify which changes could be important regarding design of the new AMR panels. The group agreed that last resort carbapenems and colistin were important targets. Amikacin was included as a marker for aminoglycoside consumption, for cross-resistance to other aminoglycosides and to improve the detection of 16S rRNA methyltransferases. No new compounds for detection of ESBL-producing bacteria were added. For *Campylobacter*, ertapenem was included to improve the detection of *ermB*.

In addition, papers have been published exploring the importance of temocillin monitoring:

J Antimicrob Chemother. 2019 Mar 1;74(3):639-644. doi: 10.1093/jac/dky493. **Evaluation of temocillin for phenotypic carbapenemase screening of *Escherichia coli* and *Salmonella enterica* isolates in relation to the presence of genes encoding ESBLs and carbapenemase production.** Lina M Cavaco, Frank Hansen, Shazad Mushtaq, Robert L R Hill, Neil Woodford, Simon Le Hello, Rene S Hendriksen, Anette M Hammerum, Henrik Hasman. PMID: 30544192 DOI: 10.1093/jac/dky493 ([Link](#))

J Antimicrob Chemother. 2014 Feb;69(2):564-7. doi: 10.1093/jac/dkt383. Epub 2013 Sep 29. **In vitro activity of temocillin against multidrug-resistant clinical isolates of *Escherichia coli*, *Klebsiella* spp. and *Enterobacter* spp., and evaluation of high-level temocillin resistance as a diagnostic marker for OXA-48 carbapenemase.** Neil Woodford 1, Rachel Pike, Daniele Meunier, Richard Loy, Robert Hill, Katie L Hopkins. PMID: 24080500 DOI: 10.1093/jac/dkt383 ([Link](#))

J Antimicrob Chemother. 2019 Dec 1;74(12):3641-3643. doi: 10.1093/jac/dkz383. **Evaluation of temocillin and meropenem MICs as diagnostic markers for OXA-48-like carbapenemases.** Katie L Hopkins, Danièle Meunier, Nazim Mustafa, Rachel Pike, Neil Woodford. PMID: 31730158 DOI: 10.1093/jac/dkz383 ([Link](#))

Summary of the plenary discussion:

It was discussed whether amikacin resistance had been detected in Gram-negative bacteria. In the chat, Denmark reported that few AMI resistant *E. coli* and *Salmonella* with MICs just above the ECOFFs, had been observed

It was discussed whether ertapenem resistant *Campylobacter* isolates had been detected. In the chat, this was reported by: Netherlands (10 *C. coli*, broilers), Belgium (few *C. coli*, cattle, MIC 1), Switzerland (few *C. jejuni*, cattle, MIC 1 or 2), Austria (one *C. coli*, MIC 1), Denmark (few *C. coli*, MIC 1).

The EURL-AR found this interesting and suggested to carry out a collaborative project on the topic for example to collect the ETP resistant *Campylobacter* isolates and carry out WGS and comparative genomics (including wild type strains). Also, it could be interesting to test the ETP resistant *Campylobacter* for resistance to other carbapenems.

EUSR (2019 data) (Pierre-Alexandre Beloeil, EFSA) See presentation ([Link](#))

Pierre-Alexandre presented the slides. AMR profiles in *Salmonella* from different animal reservoirs were presented and it was highlighted that colistin resistance is associated with particular serovars and mainly observed in broilers and laying hens. The prevalence of particular *Salmonella* serovars was associated with certain countries, animal populations and respective patterns of resistance. AMR profiles of *Campylobacter* and indicator *E. coli* were presented explaining that there is progress towards lower levels of AMR. Completely susceptible indicator *E. coli* are increasing in Europe, and there are marked differences between Northern, Eastern and Southern Europe. There is a statistically significant negative association between AMC and completely susceptible indicator *E. coli* and the prevalence of ampC/ESBL-producing *E. coli* is decreasing. MRSA isolates were reported from 4 countries in 2018 and from 7 countries in 2019.

Summary of the plenary discussion:

Decreasing trends are observed and more interventions are needed (as already indicated).

The EFSA/EURL-AR confirmatory testing (2019 data) (Jette Sejer Kjeldgaard, EURL-AR)

See presentation ([Link](#))

The confirmatory testing of 336 isolates showed that 76% were in full concordance between EURL-AR and MS according to the expected phenotype and 12 % had R/S discrepancies within the acceptable variation of the method. For the remaining 12 % of the isolates, R/S discrepancies were outside the acceptable variation of the method and the respective MS were asked to re-test the isolates and possibly correct data in the database. Altogether, data for 29 isolates were deleted or corrected in the EFSA database. The comparison of the observed geno- and phenotypes revealed $\geq 98.3\%$ of concordance. Details regarding the genotypic profiles observed for azithromycin, colistin and the ampC, ESBL and ESBL+ampC profiles were presented. No carbapenemase genes were detected.

Summary of the plenary discussion:

The categorization of isolates as ampC and/or ESBL producers were discussed. The categorization is still problematic, because it depends on the ceftiofur MIC-value (i.e. 8 versus 16 mg/L.) Beatriz Guerra (EFSA) commented that they had not yet managed to figure out how to solve this issue. A question related to the presence of *mphA* and AZI resistance was asked, and Jette explained that EURL-AR has discovered a new mechanism that soon will be published.

Survey on the application of the ESBL pre-enrichment in European laboratories (Rene S. Hendriksen, EURL-AR). See presentation ([Link](#))

The survey was conducted to understand if the NRLs would prefer alterations in the protocol for isolation and identification of ESBL/ampC/carbapenemase-producing *E. coli*, by replacing the non-selective pre-enrichment step with a selective one, which would increase specificity and sensitivity. The main point against this change is that the non-selective enrichment step allows the laboratories to use the broth on other monitoring programs, and changing it would require that they prepare new material for the other unrelated protocols. There were 34 replies and the majority of NRLs wants to keep the non-selective protocol, so at the moment, it seems that the protocol will not be altered.

Isolation Procedure for CP *E. coli* form Caeca Samples under Review (Natalie Pauly, BfR, Germany). See presentation ([Link](#))

In 2017, VIM-1 was detected in the German isolation procedure but not detected when applying the EURL-AR protocol. This non-detection lead to the question whether the isolation procedure can be approved and also about the impact of different matrix factors. I attempted to improve the isolation procedure for carbapenemase-producing *E. coli* from caeca samples as part of a work package in the One health EJP IMPART project. In the IMPART program, various modifications of the method were tested and compared to the EURL-AR method. A note to the presentation slides: In the second slide, in the timeline the green arrows correspond to Salmonella and the grey ones to *E. coli*.

The first optimization attempt was to stabilize CFU and avoid the steep decrease that occurs in the first 48 hours after collection of sample, though this was not successful. It was then attempted to include different supplementation methods both in broth and agar media and concluded that the modified procedure works best if the supplementation with antibiotics is maintained throughout the broth and agar media. However, while sensitivity, specificity and accuracy were still inferior to the EURL-AR method, selective pre-culture can be helpful to reduce overwhelming accompanying flora supported by PCR screening. The conclusion is that the best option is to use the EURL-AR method in combination with in-house prepared McC+CTX+MEM / McC +MEM (OXA-48) instead using commercial chromID agar.

Summary of the plenum discussion:

There are other projects comparing commercial and in-house media – EFSA opinion paper.

For the EURL-AR method, the window of testing from sampling has been increased from 48 hours to 96 hours. The current study underpins the need for handling the samples ASAP preferably within 24 hours. This was also what was found in IMPART.

The biggest problem appears to be the validation of the agar by the laboratories. Commercially available agar would be preferred.

The EURL-AR will consider to add an optional suggestion to supplement the media when applying the EURL-AR protocol. The media are not formally validated nor commercially available so they would have to be prepared in each NRL.

Some of the questions in the chat were left to be addressed later due to lack of time (possibly via a virtual meeting).

EFSA WGS platform for supporting AMR data collection (Mirko Rossi, EFSA)

EFSA had a discussion regarding service for supporting AMR data collection. A survey was forwarded to the MS and 13 countries responded. Eighty-five % would use a service from EFSA for extracting AMR genes. He explained that there was a survey to understand what the MS would expect to obtain from the platform. In this session, Mirko presented the slides showing the organization of the EFSA platform for AMR detection and also showed the schematic organization of the EFSA One Health WGS system and its integration with the ECDC system. He highlighted the differences between the AMR detection service and the EFSA One Health WGS system. The main goal of the joint EFSA and ECDC One Health platform is to allow data sharing for cluster detection in the context of foodborne events. While the goal of the AMR service is to provide an engine for extracting AMR genes in a format compatible with the data submission tool of EFSA.

The access of both EFSA systems will be controlled and both platforms are secure systems. Both use Nextflow as workflow manager and perform several tasks such as QC, assembly, detection of AMR genes and typing. He showed a mock example page of what it will look like and pointed out some characteristics of the system including the minimum required metadata for using the system, and that there will be limited periods of data storage.

Summary of the plenum discussion:

The system will not allow for users to submit data to EFSA, it is simply a calculation system. This system is applied if the NRL prefers to use the EFSA secure environment. There will be a test phase from 15 October 2020. The system is using ResFinder 4.1 and performs assembly and calculates QC.

The access permissions and systems are still being discussed and developed. It will be very controlled.

The AMR hits to be included in the output, are not restricted to 100% ID/COV. Details are going to be discussed later on, possibly in a webinar in the first months of 2022.

The collaboration between EFSA and ECDC “ends” at data sharing between platforms only for outbreak investigation. ECDC will probably have their own analysis pipelines.

Welcome back and introduction to the day's agenda (Rene Hendriksen, EURL-AR)

Overall outcomes of the EURL-AR EQAS 2020 for *E. coli*, *Salmonella* and *Campylobacter* (Susanne Karlsmose Pedersen, EURL-AR) See Presentation ([Link](#)).

AST of EC, SALM and CAMP and the outcome of the tests were presented. Main topics; The MIC interpretation according to EUCAST ECOFFs, ESBL categorization and species identification. A deviation was defined as an obtained result (MIC value) that differed from the expected result. To clarify what is a deviation, various examples were shown. Drug/bug combinations with deviations at a level of >25% were omitted from the analyses. Strain EC-15.7 caused some problems since the Sensititre™ plate MIC result for a NRL indicated SMX resistance whereas the expected result was SMX sensitive. EURL-AR retested the strain and confirmed a SMX sensitive phenotype. It should be mentioned that the NRL had used 20 µl inoculum whereas EURL-AR used 10 µl inoculum. In addition, strain EC-15.7 from the NRL was subjected to WGS and it revealed that the *Sul2* gene (mediates SMX resistance) had a single bp mutation at position 1. In general, data showed that the NRLs used different inoculum volumes which could have influenced the results. The result for EC-15.7/SMX was excluded from the analysis in the report.

At the laboratory level, the acceptable deviation limit has been set at 5% and for all NRLs deviation levels below this limit was observed for EC. Also for SALM strains, mainly deviations levels <5% were observed however, NRL #19 had a remarkably high deviation rate. The deviation levels for CAMP strains were very low despite two NRLs that were outliers with high deviation levels, probably due to switch of strains. ESBL phenotype interpretation: For EC some deviations and some phenotype results were correct but misinterpreted by the NRLs. For SALM, strain S-15.5 had high deviation level. Altogether, The NRLs performed very well regarding MIC testing and only minor issues were observed regarding ESBL categorization.

Challenges observed included:

A participant mentioned that strain EC-15.7 (that had caused problems regarding SMX resistance) also differed regarding MERO MIC results, when comparing result from EURL-AR and the NRL. EURL-AR has no official explanation but it was mentioned that the difference in the inoculum volume could have influenced the results. At the EURL-AR, varying MIC values have been observed when varying inoculum volumes were used, which indicates that the MIC value to some degree depend on the inoculum volume. Another participant mentioned that they also sequenced their EC-15.7 isolate and detected the *sul2* gene with a single bp mutation at position 1. Using an inoculum volume of 10µL, this NRL had observed varying MIC values for SMX (2x >1024 and 1x = 32). It was also mentioned that strain EC-15.1 had the *bla*_{CTX-M-14} gene which is characteristic for the ESBL-phenotype but the actual phenotype was found to be a carbapenemase-producer. No genetic background for the carbapenemase has been detected.

Overall outcomes of the EURL-AR EQAS 2020 for selective isolation of *E. coli* with presumptive ESBL, AmpC phenotypes or carbapenemases from meat or caecal samples (Jette Sejer Kjeldgaard, EURL-AR) See Presentation ([Link](#)).

This year's matrix included five chicken meat *E. coli* samples and three chicken caecal *E. coli* samples and was forwarded to the NRLs in Nov. 2020. It should be mentioned that DTU Food has observed that isolates from caecal samples survive better than those from other matrices. In total, 37 participants were registered and 3/33 of the participants did not isolate ESBL+ ampC phenotype from the M-6.8 caecal sample and 26/31 of the participants obtained MIC deviations regarding carbapenemase resistance for M-6.6, and this strain was omitted from evaluation. Some ESBL isolates did not present the expected phenotype, leading to a prevalence of misinterpretations: M-6.2: 55%, M-6.7: 45%, M-6.8: 50%. The acceptance criteria were changed so a dataset with a reliable sample size could be used. Overall, the result for the phenotype and the AST interpretations were good but some severe challenges were observed related to maintaining clear phenotype. Lastly, it was mentioned that the interpretive criteria are in the EQAS protocol which can be downloaded from the EURL website and that the NRLs are strongly encouraged use these.

Challenges observed included:

It was mentioned that the EURL-AR would investigate further the carbapenemase-phenotype.

It was asked by Cristina Garcia Graells if EURL-AR tests whether there are no ESBL phenotypes before spiking the samples and sending them to the NRLs and the answers was, yes the samples were tested. It was asked by Cécile Boland; what is the current recommendation for the level of inoculum volume? The EURL-AR has no official recommendation regarding the level of inoculum vol. but the latest documents from Thermo Fisher says that 10µl should be used for Gram-negatives and 30µl for Gram-positives. A NRL member also mentioned that we should harmonize the inoculum volumes and the EURL-AR replied; we will return to the network when a final decision is made.

Plenum follow-up related to EURL-AR EQAS

Approval of the three EURL-AR EQAS reports pending.

- EURL-AR EQAS *E. coli/Salmonella/Campylobacter* 2020 report
 - o Approval: The EURL-AR network were invited to bring up comments orally at the workshop or in the chat regarding the EQAS 2020 report. As no comments indicated otherwise, the report was approved and will be finalized and released by the EURL-AR.
- EURL-AR EQAS Matrix 2020 report
 - o Review and approval of the report is pending. Further information will be circulated by email.
- EURL-AR EQAS Matrix 2019 report
 - o The preliminary results were presented at the EURL-AR workshop 2020. The report is pending to be circulated for review to the network. Further information will be circulated by email.

Presentation and discussion of results and outcomes of the DTU Genomic PT 2020 - analysis of submitted sequences (Thea Kristensen, EURL-AR). See Presentation ([Link](#))

Rene explained that regarding the 2020 DTU Genomic PT results the EURL is a bit behind with the analyses and therefore, cannot results show any results at the current meeting.

In the Genomic PT 2020, 21 NRLs participated. This PT facilitated that the countries have a sufficient NGS flow in their laboratories. Two types of tests were included:

- A. Bacterial cultures (participants carry out purification of DNA and WGS).
- B. Purified DNA (the participants carry out WGS).

Sequence files (FASTQ) are uploaded to an ftp server and EURL-AR carries out trimming, assembly, QC and subsequently, a report is forwarded to the individual participants (this has been done for Genomic PT 2020).

The QC parameters were visualized in box plots and a scientific article including the procedures and results will be published.

Scoring the results showed that two laboratories were underperforming, and that generally there were difficulties or lack of results for *Campylobacter* samples. The results will be published in 2022.

Summary of the plenum discussion

Initially, it was mentioned that the GPT numbers in the slides were blinded. It was asked whether the EURL-AR in the future will consider including ENT/STAPH. The EURL-AR replied that it might be included in future PT's, but not for Genomic PT 2021, as this will include the same bacterial species as in 2020.

The observed indication of decreased performance for CAMP could be poor DNA quality or could be due to the fact that the isolates initially forwarded to the NRLs might have been contaminated.

It was asked if the evaluation report will include AMR genes. The EURL-AR responded that this will unfortunately be too demanding for the current report but a report later on could include evaluation of the AMR genes, since this is an important aspect regarding the ESBL WGS reporting for EU monitoring.

EFSA AMR ENV Scientific Opinion (Beatriz Guerra Roman, EFSA) See Presentation ([Link](#))

Rene mentioned that Beatriz G. (EFSA) suggested strengthening the One Health concept by starting environmental collaborations.

Beatriz presented the slides explaining the EFSA document "Scientific Opinion: Role played by the environment in the emergence and spread of antimicrobial resistance (AMR) through the food chain" (<https://efsa.onlinelibrary.wiley.com/doi/epdf/10.2903/j.efsa.2021.6651>).

Summary of the plenary discussion:

Rene (EURL-AR) commented that AMR in the general environment can be interpreted as a pollution issue or as a public health issue, and that interventions can focus on limiting exposure or on improving treatment. Beatriz and Rene expanded on different work from other institutions or groups that focus on particular niches of the food production chain, such as crops production and aquaculture.

Applied ECOFFs for reporting of surveillance data to the EFSA database (Pierre-Alexandre Beloeil, EFSA) See Presentation ([Link](#))

Pierre-Alexandre explained that the 2013 Commission Implementing Decision has been replaced by the 2020 issue (2020/1729/EU), going into force from January 2021. The Decision states that EUCAST ECOFFs and clinical breakpoints should be used to improve harmonization. However, there are certain antimicrobial/bacteria combinations for which no breakpoints are available and as such are not listed in the legislation. In 2021, EUCAST issued several new ECOFFs, which can be used for interpretation of results for which no breakpoint is stated in the legislation. There are also changes to breakpoints that already existed. Tables listing these alterations on a species/antimicrobial basis, also comparing both Commission Decisions (2013 and 2020), were presented.

These and other recent changes need to be taken into consideration and can be divided into:

- Values that have been used in the 2013 Decision and have changed in 2020
- Values previously recommended by EFSA and EURL-AR
- Changes made by EUCAST post-publishing of the 2020 Decision

Pierre-Alexandre provided some possible approaches such as clearly describing which breakpoints are used, if they correspond to new values compared with previous years, and what impact the previous/new breakpoints would have in the prevalence of AMR. There must also be coordination with ECDC to ensure comparability with human data.

Summary of the plenary discussion:

The Commission Implementing Decision 2020 is binding and the interpretative criteria listed in it should be followed when reporting to EFSA. For the daily testing in the laboratories, the most recent EUCAST values should be applied. These are updated continuously and probably needs frequent checks of the website to keep track of changes.

It was also discussed that historical ECOFFs were valuable at the time they were published and that we should keep historical data as they are. Rene agreed.

Thermo Scientific™ Sensititre™ EU Surveillance plates – Performance Information (Dale Clash, Thermo Scientific, UK)

Due to illness, the agenda item was cancelled.

Discussion of ideas and suggestions for upcoming EURL-AR training sessions (Jette Sejer Kjeldgaard) See Presentation ([Link](#)).

An overview of activities in previous training courses was presented. A survey was distributed in August 2021 asking for suggestions and feedback and 19 responses were received. Overall the participants were confident that there is less need for further training in phenotypic AST, and were mainly interested in receiving training in other topics like: macrolide genotypes, phylogenetic analysis and mobile genetic elements. Jette presented the interest from participants and the suggestions for the themes; i) laboratory related issues; ii) sequence analysis and genotypes; iii) WGS analysis topics. Examples of suggestions were; training on trailing endpoints in MIC reading, focus also on Nanopore sequencing, provide training on sequencing QC parameters, focus more on Gram-positive bacteria, provide theory on hybrid assemblies.

Summary of the plenary discussion:

Rene was commenting that although we do not usually focus on virulence factors, the point of including WGS is to reap the rewards of its added value. So it may be relevant to provide training on the topic, potentially by inviting experts to discuss the topic. Rene also said that past site visits have been focused on understanding workflows and identifying limitations in the NRLs, but we might be able to start providing consultations on specific issues at request from the NRLs.

It was asked if, as regards saving travel expenses and the climate impact, would it be an idea to only have physical meetings every second year and online-workshop/training the year in-between?

Response from the EURL-AR: To enhance the opportunity of networking, we currently prefer to meet once a year for training as well as for workshops.

Projects funded by EU/ECDC

Provision of EU networking and support for public health reference laboratory functions for AMR in priority healthcare-associated infections (EURGen-RefLabCap) (Anders Rhod Larsen, SSI, Denmark) See Presentation ([Link](#)).

Rene introduced these presentations and explained that they are included because it is important that the MSs are also aware of what is happening in the public health sector.

It was explained that the project is organized by the European Commission and ECDC, as a continuation of the EURGen-Net network which focuses on genotypic data from carbapenem- and/or

colistin-resistant Enterobacterales. Anders explained that the goal of EURGen-RefLabCap is to support the NRLs of countries included in the EU Health Program in their efforts to implement and/or develop genomic-based surveillance of CCRE and 2 other additional pathogens (to be determined). Anders provided an overview of the individual tasks that are needed to fulfill that goal, including establishing a reference laboratory network, review the existing capacity of NRLs and create plans for technical support based on the identified gaps and needs. He showed the link and front-page of the website (<https://www.eurgen-reflabcap.eu/>) and mentioned that the first network meeting will take place in December 2021.

Summary of the plenary discussion:

The questions and comments were postponed until after Eva's presentation.

Provision of EU networking and support for public health reference laboratory functions for AMR in *Salmonella* species and *Campylobacter* species in human samples (FWD AMR-RefLabCap) (Eva Møller Nielsen, SSI, Denmark) See Presentation ([Link](#)).

It was explained that the project is very similar to what Anders presented just before. It is also a 4 year project organized by the European Commission and ECDC but focusing on foodborne pathogens, specifically AMR on *Salmonella* and *Campylobacter*. The goals are also to improve capacity for surveillance, detection, and control of these bacteria, especially through the use of modern molecular methods such as WGS. The project includes steps equivalent to those in the EURGen-RefLabCap project, including establishing a laboratory network, developing a website with protocols and guidance documents, promoting networking between NRLs, identifying gaps in capacity and provide tailored support to priority countries. Eva showed the website (<https://www.fwdamr-reflabcap.eu/>).

Summary of the plenum discussion:

There is not necessarily an overlap between the networks, as different laboratories in the same country might be responsible for different tasks. However, if there is overlap between these new networks and other projects they are going to try to coordinate the meetings and workshops.

AOB and closing remarks (René Hendriksen, EURL-AR)

There were no further comments or questions from the participants.

Rene explained that in the future we will elaborate further on the topics:

- We will set up *ad hoc* meetings NRLs that reported ETP resistant *Campylobacter* and amikacin-resistant *Enterobacterales* to discuss this further.
- We will update the EURL-AR WGS protocol based on points raised by Mirko in his presentation

- We will set up a webinar when EFSA is ready to explain how to submit data to the platform

Any suggestions for issues to address in future EURL-AR workshops are welcome. Please send them by email to us (rshe@food.dtu.dk).

Further details related to upcoming workshop in 2022 will be circulated.
