



Work plan for 2013

The main purpose of the European Union Reference Laboratory on Antimicrobial Resistance (EURL-AR) is to ensure the quality of antimicrobial susceptibility testing in the Member States, including the use of the most optimal detection methods for antimicrobial resistant and to harmonise the procedures and methodologies used. Thus, most of the activities aim at implementing, from an analytical point of view, the provisions of monitoring of antimicrobial resistance set down in Directive 2003/99/EC of the European Parliament and of the Council of 17 November 2003 on the monitoring of zoonoses and zoonotic agents. In addition the EURL-AR provides assistance to the member States and the Commission on other relevant aspects of antimicrobial resistance. Furthermore, the EURL-AR should work in an international context and ensure that EU influences and follows global standards and guidelines.

The EURL-AR is located at the National Food Institute-DTU, as part of one of the activities of Research Group of Bacterial Genomics and Antimicrobial Resistance, within the Division of Bacterial Genomics and Epidemiology. The main activities of this research group relates to surveillance of antimicrobial resistance among bacteria from food animals, and conducting targeted research with the aim of reducing the occurrence of antimicrobial resistant bacteria among food animals and food products. The scope of activities of the Laboratory includes several international activities, education and research projects, such as the EURL-AR, the WHO collaborating center, advisory tasks, teaching, as well as several ongoing research projects (www.antimicrobialresistance.dk). The EURL-AR corresponds to circa 30.5 % of the total activities of the Research group, and to circa 12.63% of the total activities of the Division of Bacterial Genomics and Epidemiology (www.food.dtu.dk). DTU-Food is supporting (co-funding) the EURL-AR with basic housing and access to equipment, as well as administrative and IT-support.

1. Scientific advice and support to the Commission

During 2013 the EURL-AR will provide advice as stated under the general terms. The EURL-AR will participate in workshops and working groups on antimicrobial resistance initiated by EFSA, EMA, ECDC, Codex, FAO/WHO/OIE and when relevant other organisations. The WHO has established an Advisory Group in Surveillance of Antimicrobial Resistance (AGISAR), which has as the aim to develop global standards for monitoring of antimicrobial resistance. The EURL-AR is obliged to actively support this initiative.

For 2013 the EURL-AR plans to take part in several activities and the budgeted workload is expected to represent around 6,5% of the total EURL-AR salary expenses. Travel expenses related to this activity are calculated to circa 3,150 EUR, for 3-4 short travels including 1 or 2 members of the EURL-AR staff.

2. Co-ordination of National Reference Laboratories and provision of technical support

a. Meetings on standardization of monitoring of antimicrobial resistance

An important problem in relation to ring trials and monitoring of resistance is the lack of common interpretive criteria. This is a global problem and not only related to EU. The EU is the world's largest exporter and importer of food products and European citizen travel with an

increasing frequency outside the EU. Thus, international collaboration and the development and harmonisation of global standards is a high priority, as also indicated in the Commission's strategy for antimicrobial resistance. The EURL-AR will in 2013 continue the work with WHO (AGISAR) and other important stakeholders such as ECDC, EUCAST, CLSI, OIE, FAO and Global Foodborne Infections Network (GFN) in order to promote a common international standard for harmonization of antimicrobial resistance monitoring and support of capacity building in member countries for antimicrobial resistance monitoring.

b. Maintaining the network of NRL's

The EURL-AR will during 2013 maintain and continuously update a full list of contact persons from all NRL's. In addition, the EURL-AR will attempt to identify expected members from applicant countries to include in the network. This list will also be maintained during the following years.

c. Dissemination of knowledge and information

The EURL-AR will maintain the EURL-AR web page (www.eurl-ar.eu) where relevant information is posted. In addition, the EURL-AR will distribute updates, highlights or other relevant information through newsletters to the NRL's. Specifically for 2013 the EURL-AR will provide updates on the new tools as regards for example the genotypic characterisation of resistance genes and transferable elements carrying resistance genes.

The EURL-AR will provide updated lists of suggested cut-off values based on the work done by EUCAST (www.eucast.org) and other international standardization committees. Especially the EURL-AR will engage in activities with EFSA and ECDC, aiming towards globally standardised breakpoints and cut-off values as mentioned above. In addition, the latest guidelines on which antibiotics to test for and ranges to use as well as other problems encountered will also be disseminated between the NRL's.

The EURL-AR will continue to disseminate latest state of the art information as regards scientific findings. This will include emerging resistance issues related to for example resistance mechanisms or genes in relevant microorganisms.

The technological developments in whole genome sequencing will soon allow this to be used in routine diagnostic, either as a supplement or a replacement to currently used phenotypic techniques. The EURL-AR will continuously keep the network closely informed about developments in this area.

d. Collection of data of methodology and plate designs used for the performance of AST testing, using commercial microtiterplates in the network.

The golden standard method for antimicrobial susceptibility testing (AST) is the minimum inhibitory concentration (MIC) testing methodology which has been adopted in most MS for harmonization purposes and to improve the quality of susceptibility testing in general. However, there are different brands and plate designs available, different methodologies and equipment which can be used in the performance of such tests. We intend to utilize the information collected in 2012 (questionnaire-based survey) for trouble shooting in connection with the results obtained in proficiency tests as well as for evaluation and revision of MIC-panel designs in order for AST results to comply with current guidelines.

e. Improve and extend databases of primers, reference material and antimicrobial resistance genes.

For detection and characterization of resistance mechanisms it is necessary to identify an extremely large number of resistance genes and mutations responsible for the phenotypes observed. At the EURL-AR, databases containing primers and reference strains are currently available to our network for setting up molecular methods for detection. In addition, a database/tool for identification of most known genes and variants based on sequence data (including reads and assembled whole genomes) is available (ResFinder). These databases will continuously be improved and extended. All EU agencies will have free access to and can obtain a copy of the database. In 2012 we have started to collect some information on available sequences, and this will be continued in 2013.

In 2013 the EURL-AR intends to continue the above mentioned activities (a, b, c, d and e) which represent circa 14,4% of the total workload of the EURL-AR. Also under this activity there is a need to use updated MAC-computers which allow access to non-windows based software platforms for the tasks required, therefore we include the expenses relatively to one computer acquired in 2011 and expect to purchase another computer in the period of 2013, as included in the budget. Furthermore, we expect to acquire a new stand alone nephelometer for inoculum adjustment in antimicrobial susceptibility testing.

3. Ring trials, comparative testing and quality assurance

External quality control is an important part of ensuring and maintaining the analytic quality of laboratory tests performed. The EURL-AR will in the spring and autumn 2013 organize the following ring trials on antimicrobial susceptibility testing for the NRL's:

- a. *Salmonella*
- b. *Campylobacter*
- c. *Escherichia coli*
- d. Enterococci
- e. Staphylococci
- f. Detection of MRSA
- g. Genotypic characterization of relevant antimicrobial resistant strains including detection of ESBL-genes (optional)

The organization and evaluation of the results are given under the general terms.

The EURL-AR will provide protocols for speciation of *Enterococcus* and *Campylobacter* in relation to identification of the strains sent out for the proficiency test.

The ECDC-network of Food and Waterborne Diseases will be offered the participation in the *Salmonella* and *Campylobacter* proficiency test. The expenses for this will be fully covered by the ECDC-network.

Currently, working groups are working on updated recommendations for monitoring and suggestions for baseline studies. The EURL-AR will as far as possible adjust the ring trials so they fit any future suggestions and thereby continue to fulfil the objectives of Directive 2003/99/EC.

The activities related to the organization of the ring trials, shipments, evaluation of results, production of reports and the maintenance of a quality assurance system with accredited proficiency testing encompasses the largest part of the EURL-AR staff workload, involving

both academic personnel (Quality assurance, organization, communication with NRL's, planning, data collection, data analysis and report writing and presentation of results, follow up on deviations and /or missions related to follow up on ring trial results) and technicians (testing of isolates, preparation of samples, quality control activities, shipping of samples) representing circa 55,4% of the total salary expenses. Furthermore, the organization of ring trials represents circa 90% of the consumables and 90% of the shipping expenses.

4. Evaluation and development of analytic methods

Reference strains

Reference strains for use in quality control or other analyses are an important part of the internal quality control and validation of on-going analyses. The EURL-AR will continuously extend its already available strain collection and make the strains available for NRL's on request.

Interpretative criteria

The EURL-AR will if needed perform studies on the susceptibility of food borne pathogens to various antimicrobial agents in order to provide data for the establishment of interpretative criteria for categorizing isolates as susceptible or resistant. On the annual work shop between all NRL's the most urgent needs and problems were discussed. Thus, especially for 2013 the EURL-AR will initiate the following projects:

- As follow up of the survey on MIC methods and plate design there might be the need to perform comparisons of the performance of different plates under the same conditions.
- The EURL-AR will facilitate the collection and dissemination of antimicrobial susceptibility data of relevant veterinary and food microorganisms for EUCAST.
- Determine the phenotypic MIC value based on single gene cloning focusing on carbapenemase-genes.
- Detection of resistance mechanisms in *Campylobacter* for implementation in the Resfinder.
- Correlate observed presence of resistance genes to phenotypes

MRSA detection

The emergence of MRSA in food animal production is still a matter of concern. The EURL-AR will together with other institutions continue to evaluate different methods for the optimal detection of MRSA from animal sources and if possible food of animal origin. Furthermore, the EURL-AR will follow the recent findings and implement tools for characterization of new emerging types and/or genetic elements.

Extended spectrum beta-lactamases

The emergence of ESC producing isolates poses a major problem for human health. The EURL-AR has together with the NRL's collected information on the occurrences of ESC resistance in Europe and will in 2013 summarize and publish the findings. The EURL-AR will in addition to supporting activities for EFSA, perform method testing to evaluate semi-quantitative methods and evaluate methods for detection of carbapenem resistance.

The activities related to evaluation and development of analytical methods will involve close collaboration between elements of the EURL-AR and of the NRLs, representing therefore excellent networking opportunities. The workload related to these projects is expected to represent circa 11,5% of the salary expenses for the EURL-AR in 2013, 7% of the

consumables and a 10% of the shipping costs due to the need of exchange of materials, isolates or reference strains between the involved laboratories.

5. Confirmatory testing

The EURL-AR will provide confirmatory testing for NRL's on bacterial isolates of particular relevance or on request by the European Commission. Specifically, the EURL-AR will provide reference testing of putative *Salmonella* and *E. coli* isolates producing beta-lactamases with extended spectrum, and carbapenemases. Additionally, the EURL-AR will also provide reference testing to the NRLs for characterization of isolates resistant to fluoroquinolone or harbouring transferable fluoroquinolone resistance mechanisms, and confirmation of MRSA.

Confirmatory testing requests are sent on ad-hoc basis and their volume for 2013 is therefore not predictable, depending on the needs of the Commission, and the NRLs, we have therefore calculated that these represent circa 5% of the salary expenses and 3% of the consumables used in the laboratory.

6. Missions for specific assistance to individual laboratories, site visits, individual training courses, or conferences

Some NRL's might have a need for special assistance. The EURL-AR will to the extent possible within the financial limits provide specific assistance to individual laboratories based on individual needs in the follow up of ring trial results or need to implement methodology.

In 2013 the EURL-AR plans to visits selected NRLs that require special training. The organization of individual training programmes including hand-on practical training is expected to represent circa 4,5% of the EURL-AR workload and additional mission travel expenses are calculated to 7350 EUR, for three 4 to 5-day travels including 2 members of EURL-AR staff.

7. E-learning

The continuous changing of staff at the different NRL's make it difficult to ensure sufficient training through individual and larger training courses. The EURL-AR will increase the focus on this point and invest more efforts in creation of e-learning tools during the next years. These activities will be initiated in 2013, by starting the preparation of specific learning materials to be included in interactive interfaces for e-learning. The first focus will be on the creation of tools to teach the most basic principles and methods of susceptibility testing, and detection of emerging resistance problems. The creation of the informatics platforms and development of the tools is postponed to 2014.

This activity is expected to represent circa 2,6% of the total EURL-AR workload for 2013.

8. Workshop

An integrated approach is planned for the EURL-AR workshop in 2013. The meeting schedule will include a joint day between the ECDC-FWD-network and the EURL-AR network. The meeting venue will be DTU, Denmark. All expenses and administration related to hosting the ECDC-FWD network will either be covered by ECDC or as in kind by DTU. EU funding will solely be used to host the EURL network. The agenda will include the following key components:

- Update from EURL-AR, EFSA, ECDC, the European Commission and other parties
- Results of the proficiency tests performed in 2012
- Integrated monitoring aspects
- Presentation of scientific projects and other activities at the NRL's

The 2013 workshop will include participants from the NRL network, invited speakers from relevant organisations and representatives of non-EU countries (up to a 45 participants in total according to the regulation). This will hopefully give opportunity to expand the network and exert more influence on the quality of susceptibility testing performed in non-EU countries.

9. Training course

The EURL-AR plan to arrange a training course for 32 participants of the NRL's focused on harmonization, assuring that all MS are able to perform the harmonized monitoring according to the new legislation that will apply from 2014. This includes identification and phenotypical testing of ESBL and MRSA as well as specific needs in terms of basic antimicrobial susceptibility testing.

The 2013 training course will include participants from the NRL network, invited speakers and representatives of non-EU countries.