Work plan for 2012

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 The Research Group for Antimicrobial Resistance and Molecular Epidemiology within the Division for Microbiology and Risk Assessment. 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 17% of the total activities of the Research group, and to circa 4,8% of the total activities of the Division for Microbiology and Risk Assessment (www.food.dtu.dk). DTU-Food is supporting (co-funding) the EURL-AR with providing basic housing and access to equipment, as well as administrative and IT-support.

1. Scientific advice and support to the Commission

During 2012 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, 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 that also involves EFSA.

For 2012 the EURL-AR plans to take part in several activities and the budgeted workload is expected to represent around 5% of the total EURL-AR salary expenses. Travel expenses related to this activity are calculated to circa 3000 Euro, 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

The most 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, the development and harmonisation of global standards is a high priority. The EURL will in 2012 continue the work with WHO (AGISAR)

and other important stakeholders such as 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 will during 2012 maintain and continuously update a full list of contact persons from all NRL's. In addition, the EURL 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 will maintain the EURL web page (www.eurl-ar.eu) where relevant information is posted. In addition, the EURL will distribute updates, highlights or other relevant information through newsletters to the NRL's. Specifically for 2012 the EURL-AR will provide updates on the current situation in Europe on extended spectrum beta-lactamase producing and quinolone resistant bacteria, as well as methicillin-resistant *Staphylococcus aureus* and other issues considered critically important.

The EURL-AR will provide updated lists of suggested cut-offs based on the work done by EUCAST (www.eucast.org) and other international standardization committees. Specially the EURL-AR will host and participate in meetings with the aim to get standardised breakpoints and cut-off values globally as mentioned above. In addition, knowledge 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 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 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 start to collect information using a questionnaire-based survey on panel designs used, as well as methods, media and reagents used in each laboratory. This will allow cooperation between labs for acquisition of plates, exchange of methods, and to obtain information about the methods/ressources available in each individual laboratory. This information will be compiled during the start of 2012 and analysed and the results are expected to be presented at the EURL-AR Workshop.

e. Construction of a database of 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 we already maintain a databases containing primers and reference strains which are available to our network for setting up molecular methods for detection.

However, a database including all known genes and variants with the respective DNA sequences is not available so far. Furthermore, the current database is not directly accessible for the different member states. We will update this database and make the information available for the different NRL's, We expect the data collected in this database to be made available to the network and to be used for implementing new tools for detection of resistance genes in the MS (for example from sequenced genomes, or in the simple design of primers for PCR methods) furthermore all EU agencies will have free access to and can obtain a copy of the database. In 2011 we have started to collect some information on available sequences, and this will be continued in 2012.

In 2012 the EURL-AR intends to continue the above mentioned activities (a,b, c, d and e) which represent circa 15% 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 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 2012, as included in the budget.

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 2012 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 Gram positive and Gram negative bacteria (optional)

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

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% 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

<u>Reference strains</u>

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 2012 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.
- As discussed at the workshop, the EURL-AR will collaborate with some NRL's to Investigate colistin resistance in *Salmonella* strains from certain serovars which display colistin MIC= 4mg/L.
- Pilot evaluation of using genetic detection as a supplement to phenotypic testing.

MRSA detection

The emergence of MRSA in food animal production is a matter of increasing 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, as there are still some issues with sensitivity of methods for detection. 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 2012 summarize and publish the findings.

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 10% of the salary expenses for the EURL-AR in 2012, 6,5% of the consumables and a minor part 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 2012 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 3,5% of the consumables used in the laboratory.

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

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 2012 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 5% of the EURL-AR workload and additional mission travel expenses are calculated to 7000 Euro, 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 though 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. Therefore, more activities will be set up in 2012, for the implementation of learning tools and 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.

As referred before this activity is budgeted separately from the other activities related to the support of NRL's as it will increase in focus and is expected to represent circa 5% of the total EURL-AR workload for 2012 and including mostly work on tool design and development.

Workshops

In 2012, the EURL-AR will host a workshop held in Kgs. Lyngby, Denmark with the following tentative agenda:

- Introduction and presentation
- Update of the 2011 activities and functions of the EURL-AR
- Update on other EURL's
- Update from EFSA, the European Commission and other parties
- Updates on ongoing projects
- Update on collection of data on MIC methods.
- Break points and cut-off values, updates and new projects
- Results of the ring trials performed in 2011
- Presentation and discussions on national programmes on susceptibility testing.
- Presentation of activities at the NRL's
- Experiences from non EU countries

The 2012 workshop will include participants from the NRL network, invited speakers from relevant organisms and representatives of non-EU countries which according to the new EU regulation, may be covered by the EC funding. This will hopefully give opportunity to expand the network exert more influence on the quality of susceptibility testing non-EU countries.

The expected expenses for the forthcoming 2012 workshop were therefore calculated for a maximum of 45 participants.