

Confirmatory testing in relation to reporting antimicrobial resistance in bacteria from animals and food



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DTU Food National Food Institute

Term of Reference, EURL-AR



Confirmatory testing

- The EURL-AR will provide confirmatory testing on bacterial isolates of particular relevance upon request by the NRLs, the European Commission and EFSA:
 - This specific targeted confirmatory testing will be offered by the EURL-AR to support the activities related to the implementation of Decision 2013/652/EU
 - This confirmatory testing will be used to assure the quality of results submitted by MSs, to detect resistance mechanisms and to determine correspondence between phenotypes and genotypes
 - The EURL-AR will offer Material Transfer Agreements (MTAs) between the EURL-AR and MSs to define the legal aspects of strain and data ownership

Work-flow



Upon reception of isolates at EURL-AR:

- MIC determination on EUVSEC and EUVSEC 2
- WGS (Illumina MiSeq, Illumina HiSeq)
- Data analysis: assembly, batch upload (CGE), correspondence phenotype-genotype
- Communication with MSs:
 - troubleshooting
 - submission to ENA (only upon permission from MSs)

Overview (MON. 2014)



- The isolates were received between December 2015 and February 2016
- 83 isolates were tested on the EUVSEC panel
- 100 isolates were tested on the EUVSEC2 panel
- 165 WGS of which 141 were analyzed using the CGE tools (ResFinder, MLST, Plasmid Finder, etc.)
- 8 isolates from 5 countries were omitted due to contaminations
- 13 isolates from 5 countries were not received
- Phylogenetic analysis (31 S. Infantis and 18 S. Kentucky strains)
- Data sent to MSs in August 2016

Overview (MON. 2015)



- The isolates were received between August and December 2016
- 210 isolates requested
 - 9 isolates were not received
 - > 11 isolates arrived in mixed cultures that had to be excluded
 - ➤ 190 isolates were tested on EUVSEC and EUVSEC 2
- MIC data reported to MSs in December 2016
- WGS data available only recently, ongoing analysis

Correspondence phenotype-genotype: intepretation of the genotypic results

Color codes GENOTYPE									
Agreement phenotype-genotype at both labs (if data from only one lab are provided, this is indicated in the comments field)									
Agreement phenotype-genotype at EURL only									
Agreement phenotype-genotype at MS only									
The gene detected does not explain (fully) the phenotype									
Not detected phenotypically (antimicrobial not in the panel/panel not tested)									
In parenthesis, next to each gene, there is a value showing if the hit is partial or has less than 100% identity to the reference gene									
Incorrect serovar identificatio	n								



Follow-up:

MICs and correspondence genotype-phenotype

- At MSs: self-evaluation
- At EURL-AR:

Peculiar cases:

- Phenotypic resistance but no genes detected (gentamicin, trimethoprim, ampicillin, cefoxitin, cefotaxime, ceftazidime, colistin)
- Ertapenem-resistant CMY-2-producing *E. coli*: porin deficiency? Specific mutations?
- Contact MS EURL-AR:

Problems identified:

- ➤ contaminations
- plasmid finder results are different from those from the PBRT

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Thank you for your attention

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