8-9 October 2024

18th EURL-AR Workshop

EFSA AMR on-going activities

Raquel García-Fierro

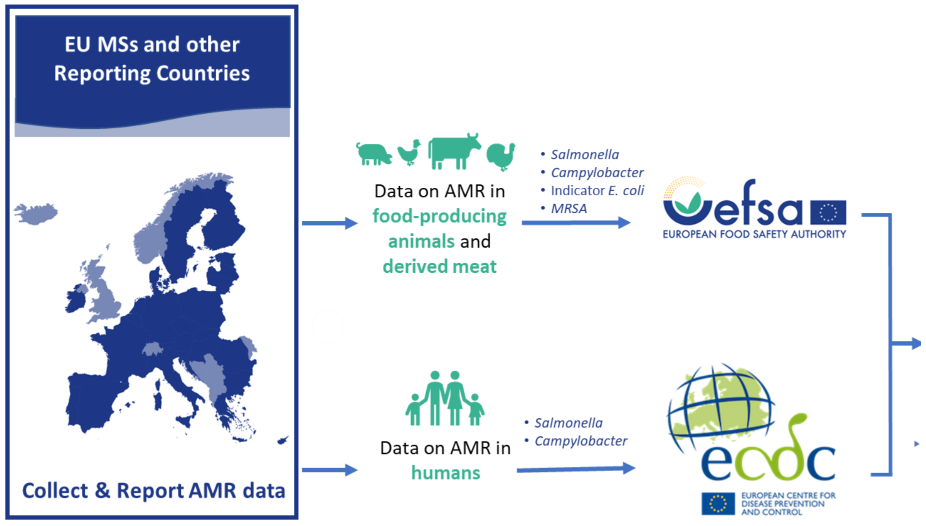
EUROPEAN FOOD SAFETY AUTHORITY

AMR monitoring

AMR MONITORING – 2022-2023 EUSR ON AMR

Commission Implementing Decision 2020/1729/EU

Lays down specific tech. requirements **2021 - 2027**


EU MSs and other Reporting Countries

Collect & Report AMR data

Data on AMR in **food-producing animals** and **derived meat**

- *Salmonella*
- *Campylobacter*
- Indicator *E. coli*
- *MRSA*

Data on AMR in **humans**

- *Salmonella*
- *Campylobacter*

2021-2022 EUSR on AMR


EU Summary Report on Antimicrobial resistance

Approved: 19 January 2024

DOI: 10.2903/j.efsa.2024.8583

SCIENTIFIC REPORT

The European Union summary report on antimicrobial resistance in zoonotic and indicator bacteria from humans, animals and food in 2021–2022

European Food Safety Authority (EFSA) | European Centre for Disease Prevention and Control (ECDC)

Correspondence: zoonoses@efsa.europa.eu (EFSA) and fwd@ecdc.europa.eu (ECDC)

Abstract

This report by the European Food Safety Authority and the European Centre for Disease prevention and Control, provides an overview of the main findings of the 2021–2022 harmonised Antimicrobial Resistance (AMR) monitoring in *Salmonella* spp., *Campylobacter jejuni* and *C. coli* from humans and food-producing animals (broilers, laying hens and fattening turkeys, fattening pigs and cattle under one year of age) and relevant meat thereof. For animals and meat thereof, AMR data on indicator commensal *Escherichia coli*, presumptive extended-spectrum beta-lactamases (ESBL)-/AmpC beta-lactamases (AmpC)–/carbapenemase (CP)-producing *E. coli*, and the occurrence of methicillin-resistant *Staphylococcus aureus* (MRSA) are also analysed. Generally, resistance levels differed greatly between reporting countries and antimicrobials. Resistance to commonly used antimicrobials was frequently found in *Salmonella* and *Campylobacter* isolates from humans and animals. In humans, increasing trends in resistance to one of two critically antimicrobials (CIA) for treatment was observed in poultry-associated *Salmonella* serovars and *Campylobacter*, in at least half of the reporting countries. Combined resistance to CIA was however observed at low levels except in some *Salmonella* serovars and in *C. coli* from humans and animals in some countries. While CP-producing *Salmonella* isolates were not detected in animals in 2021–2022, nor in 2021 for human cases, in 2022 five human cases of CP-producing *Salmonella* were reported (four harbouring bla_{OXA-48} or $bla_{OXA-48-like}$ genes). The reporting of a number of CP-producing *E. coli* isolates (harbouring bla_{OXA-48}, $bla_{OXA-181}$, bla_{NDM-5} and bla_{VIM-1} genes) in fattening pigs, cattle under 1 year of age, poultry and meat thereof by a limited number of MSs (5) in 2021 and 2022, requires a thorough follow-up. The temporal trend analyses in both key outcome indicators (rate of complete susceptibility and prevalence of ESBL-/AmpC-producers in *E. coli*) showed an encouraging progress in reducing AMR in food-producing animals in several EU MSs over the last 7 years.

KEYWORDS

antimicrobial resistance, ESBL, indicator bacteria, MRSA, zoonotic bacteria

- Annual report
- Jointly produced by EFSA and ECDC
- Summarises AMR findings for two consecutive years

https://doi.org/10.2903/j.efsa.2024.8583

Publication date: 28th of February 2024

✓ Outcomes of the monitoring

- **Occurrence of AMR**
- Combined resistance to **hp-Critically Important Antimicrobials (CIAs)**
- **Key Outcome Indicators** of AMR:
  - **Complete Susceptibility** in indicator *E. coli*
  - **Prevalence of ESBL/AmpC- producers**
- **Temporal trends**

2022- 2023 EUSR on AMR

2022 AMR data: poultry populations

2023 AMR data: pigs and calves

Timelines:

Currently under production

Consultation: December 2024

Expected Publication date: February 2025

AMR MONITORING – CONFIRMATORTY TESTING EXERCISE

Reference testing exercise that includes:

- Confirmatory testing of the antimicrobial susceptibility and
- Whole genome sequencing (WGS) analysis of the selected isolates

2023 conf. testing exercise

A total of 328 isolates selected

- Indicator *E.coli: 268*
- *Campylobacter: 52*
- *Salmonella: 5 isolates*

Reporting period for 2023 zoonoses, FBO and AMR data: timelines

1 March
Data collection OPEN

31 May
Data collection CLOSED

01 – 12 June
First Validation Period (EFSA)
Data collection CLOSED

12 June Sent out
First Validation Letter to MSs
Data collection OPEN

12 – 27 June
First Correction Period (MS)
Data collection OPEN

28 June – 9 July
Final Validation Period (EFSA)
Data collection CLOSED

9 July Sent out
Second Validation Letter to MSs
Data collection OPEN

9 - 17 July
Final last correction period (MS)
Data collection OPEN

19 July
EFSA uses DATA from DWH dated 19 July 2024 to produce EUSR 2023
Data collection CLOSED

1 – 30 November
Amendment of 2023 and historical data
Data collection OPEN

Confirmatory testing exercise timelines:

8 July: EFSA sends to the EURL-AR the list of isolates for the confirmatory testing exercise + MIC results

9 July: EURL-AR sends the emails to the MSs requesting the selected isolates

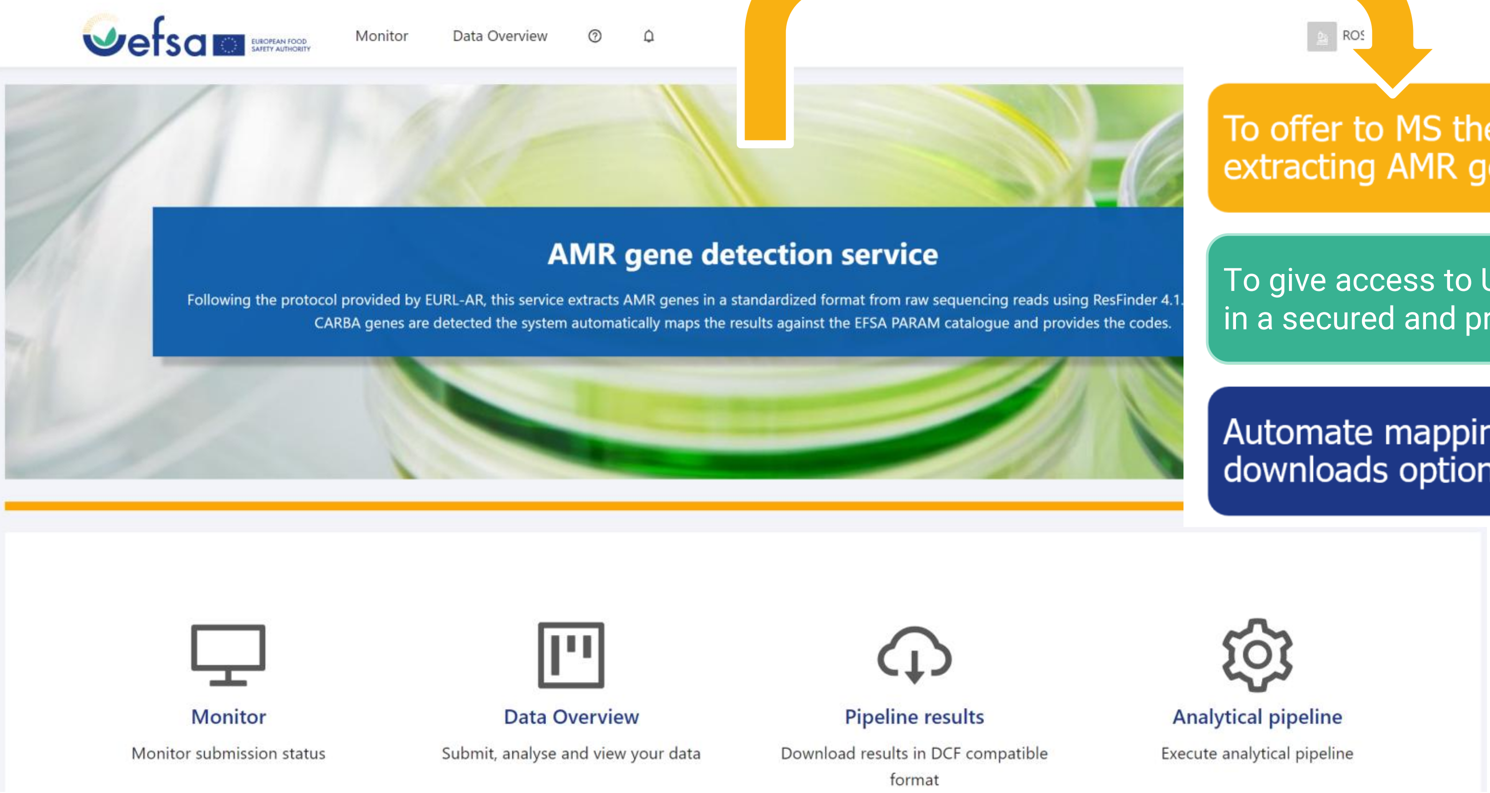
1 September: All requested isolates should arrive to the EURL-AR

14-15 October: Finalisation of the confirmatory testing exercise and starting the comparison of results

31 October: EURL-AR sends the results of the confirmatory testing exercise to the MSs

Icons from www.flaticon.com

THE EFSA AMR GENE DETECTION SERVICE



AMR gene detection service

Following the protocol provided by EURL-AR, this service extracts AMR genes in a standardized format from raw sequencing reads using ResFinder 4.1. CARBA genes are detected the system automatically maps the results against the EFSA PARAM catalogue and provides the codes.

To offer to MS the opportunity to use EFSA computing resources for extracting AMR genes from WGS data

To give access to User-Friendly interface for accessing and managing the data in a secured and private environment, no sharing of data

Automate mapping against the PARAM catalogue and different downloads options for the results

ResFinder application: v. 4.4.2 (2023-11-27)
ResFinder database: v. 2.2.1 (2023-10-27)
PointFinder database: v. 4.0.1 (2023-11-02)
DisinFinder database: v. 2.0.1 (2023-05-31)

The service is available for

Salmonella enterica
Escherichia coli
Campylobacter jejuni
Campylobacter coli

For access, please send email to
mirko.rossi@efsa.europa.eu

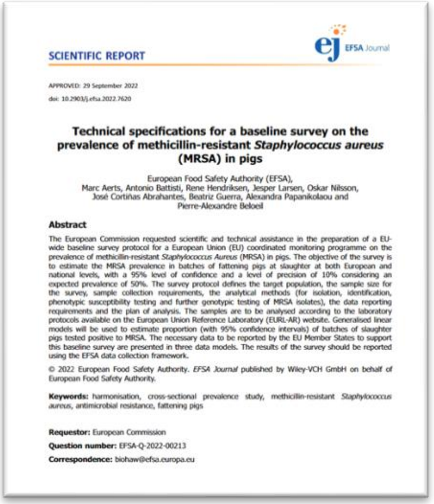
UP-COMING EU-WIDE BASELINE SURVEYS ON AMR

MRSA in pigs

- To assess the **prevalence** of MRSA
- To assess the **AMR profile**
- To assess the **genetic diversity**
- To be performed in **2025**

AMR in bacteria from aquaculture animals

- **Mandate** of the European Commission (2023)
- EFSA Expert Working Group
  -> **EFSA Technical Specifications** (July 2024)



SCIENTIFIC REPORT

EFSA Journal

APPROVED: 29 September 2022

doi: 10.2903/j.efsa.2022.7620

Technical specifications for a baseline survey on the prevalence of methicillin-resistant *Staphylococcus aureus* (MRSA) in pigs

European Food Safety Authority (EFSA),
Marc Aerts, Antonio Battisti, Rene Hendriksen, Jesper Larsen, Oskar Nilsson,
José Cortiñas Abrahantes, Beatriz Guerra, Alexandra Papanikolaou and
Pierre-Alexandre Beloeil

Abstract

The European Commission requested scientific and technical assistance in the preparation of a EU-wide baseline survey protocol for a European Union (EU) coordinated monitoring programme on the prevalence of methicillin-resistant *Staphylococcus Aureus* (MRSA) in pigs. The objective of the survey is to estimate the MRSA prevalence in batches of fattening pigs at slaughter at both European and national levels, with a 95% level of confidence and a level of precision of 10% considering an expected prevalence of 50%. The survey protocol defines the target population, the sample size for the survey, sample collection requirements, the analytical methods (for isolation, identification, phenotypic susceptibility testing and further genotypic testing of MRSA isolates), the data reporting requirements and the plan of analysis. The samples are to be analysed according to the laboratory protocols available on the European Union Reference Laboratory (EURL-AR) website. Generalised linear models will be used to estimate proportion (with 95% confidence intervals) of batches of slaughter pigs tested positive to MRSA. The necessary data to be reported by the EU Member States to support this baseline survey are presented in three data models. The results of the survey should be reported using the EFSA data collection framework.

© 2022 European Food Safety Authority. *EFSA Journal* published by Wiley-VCH GmbH on behalf of European Food Safety Authority.

Keywords: harmonisation, cross-sectional prevalence study, methicillin-resistant *Staphylococcus aureus*, antimicrobial resistance, fattening pigs

Requestor: European Commission

Question number: EFSA-Q-2022-00213

Correspondence: biohaw@efsa.europa.eu

https://www.efsa.europa.eu/en/efsajournal/pub/7620

L 136/78 EN Official Journal of the European Union 24.5.2023

COMMISSION IMPLEMENTING DECISION (EU) 2023/1017

of 23 May 2023

amending Implementing Decision (EU) 2020/1729 as regards the monitoring of methicillin-resistant *Staphylococcus aureus* (MRSA) in fattening pigs

(notified under document C(2023)3251)

(Text with EEA relevance)

THE EUROPEAN COMMISSION,

Having regard to the Treaty on the Functioning of the European Union,

Having regard to Directive 2003/99/EC of the European Parliament and of the Council of 17 November 2003 on the monitoring of zoonoses and zoonotic agents, amending Council Decision 90/424/EEC and repealing Council Directive 92/117/EEC (1), and in particular Articles 4(5), 7(3) and Article 9(1), fourth subparagraph, thereof,

Whereas:

Commission Implementing Decision 2023/1017/EU

Lays down specific tech. requirements

Approved: 2 July 2024

DOI: 10.2903/j.efsa.2024.8928

SCIENTIFIC REPORT

efsa JOURNAL

Technical specifications for a EU-wide baseline survey of antimicrobial resistance in bacteria from aquaculture animals

European Food Safety Authority (EFSA) | Marc Aerts | Sandrine Baron | Valeria Bortolaia | Rene Hendriksen | Beatriz Guerra | Anca Stoicescu | Pierre-Alexandre Beloeil

Correspondence: biohaw@efsa.europa.eu

Abstract

The European Commission requested scientific and technical assistance in the preparation of a EU-wide baseline survey of antimicrobial resistance (AMR) in bacteria from aquaculture animals. It is recommended that the survey would aim at estimating the occurrence of AMR in *Aeromonas* spp. isolated from Atlantic Salmon (*Salmo salar*), European seabass (*Dicentrarchus labrax*) and trout (*Salmo trutta, Salvelinus fontinalis, Oncorhynchus mykiss*) intended to consumption, at harvesting (at farm/slaughter), at the EU level and in addition, at estimating the occurrence and diversity of AMR of *Escherichia coli*, *Enterococcus faecium*, *Enterococcus faecalis*, *Vibrio parahaemolyticus* and *Vibrio alginolyticus* in blue mussel (*Mytilus edulis*) and Mediterranean mussel (*Mytilus galloprovincialis*) from production areas and at dispatch centres at the EU level. These technical specifications define the target populations, the sample size for the survey, sample collection requirements, the analytical methods (for isolation, identification, phenotypic susceptibility testing and further genotypic analysis of some of the bacteria targeted) and the data reporting requirements. The data to be reported by the EU Member States to support this baseline survey are presented in three data models. The results of the survey should be reported using the EFSA reporting system.

KEYWORDS

antimicrobial resistance, aquaculture, cross-sectional prevalence study, finfish, mussels

https://www.efsa.europa.eu/en/efsajournal/pub/8928

Joint reporting on the consumption of antimicrobial agents and occurrence of AMR in bacteria from humans and food-producing animals (JIACRA)

- As part of the implementation of the EU action plans on AMR, the **European Center for Disease Prevention and Control** (ECDC), the **European Food Safety Authority** (EFSA) and the **European Medicines Agency** (EMA) have been requested by the European Commission to analyse possible **relationships** between the **consumption of antimicrobial agents** and the **occurrence of antimicrobial resistance** in humans and food-producing animals.

- JIACRA builds on previous collaboration between the three EU agencies, including harmonisation of approaches, expertise and previous joint publications on related subjects.

- So far, four JIACRA reports have been published, with a fifth report expected to be requested later this year.

2015
(2011-2012)

2017
(2013-2015)

2021
(2016-2018)

2024
(2019-2021)

2026?

AMR Scientific Opinions

BIOHAZ MANDATE - VIBRIO IN SEAFOOD IN THE EU – PUBLISHED

The BIOHAZ Panel is asked to issue a scientific opinion on the **public health aspects of *Vibrio parahaemolyticus*, *Vibrio vulnificus* and non-O1, non-O139 *Vibrio cholerae*** (and other species whenever relevant) **related to the consumption of seafood for the EU population**:

- **ToR1.** To review, for the **relevant *Vibrio* spp.**, the existing information on **occurrence and concentration** in seafood, available analytical **methods**, **pathogenicity** to humans and virulence factors, as well as **antimicrobial resistance and persistence mechanisms** in different environments.
- **ToR2.** To identify the **factors** in the aquatic environments and in food (including during production and processing) that influence occurrence and growth of the relevant *Vibrio* spp., and **affect transmission of their virulence and resistance determinants**.
- **ToR 3 to 6 ….**

Published: **23-07-2024**

Adopted: 12 June 2024

DOI: 10.2903/j.efsa.2024.8896

efsa JOURNAL

SCIENTIFIC OPINION

Public health aspects of *Vibrio* spp. related to the consumption of seafood in the EU

EFSA Panel on Biological Hazards (BIOHAZ) | Konstantinos Koutsoumanis | Ana Allende | Avelino Alvarez-Ordóñez | Declan Bolton | Sara Bover-Cid | Marianne Chemaly | Alessandra De Cesare | Lieve Herman | Friederike Hilbert | Roland Lindqvist | Maarten Nauta | Romolo Nonno | Luisa Peixe | Giuseppe Ru | Marion Simmons | Panagiotis Skandamis | Craig Baker-Austin | Dominique Hervio-Heath | Jaime Martinez-Urtaza | Eva Sanjuán Caro | Eckhard Strauch | Anne Thébault | Beatriz Guerra | Winy Messens | Ancuta Cezara Simon | Rubén Barcia-Cruz | Elisabetta Suffredini

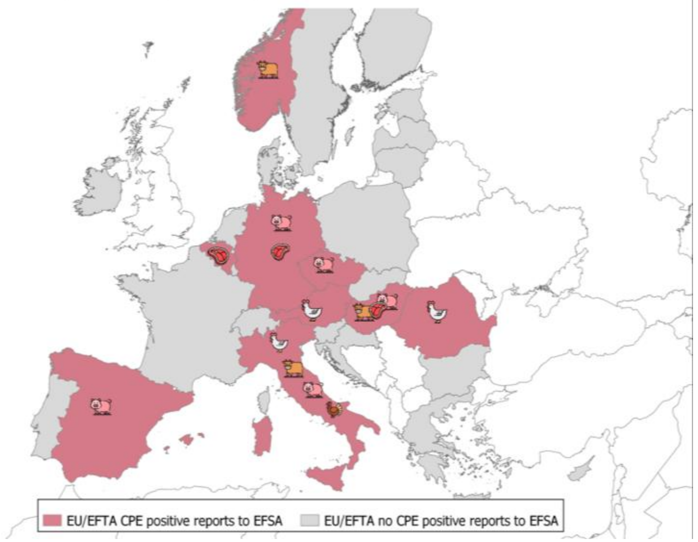
Correspondence: biohaz@efsa.europa.eu

Abstract

Vibrio parahaemolyticus, *Vibrio vulnificus* and non-O1/non-O139 *Vibrio cholerae* are the *Vibrio* spp. of highest relevance for public health in the EU through seafood consumption. Infection with *V. parahaemolyticus* is associated with the haemo-

https://efsa.onlinelibrary.wiley.com/doi/epdf/10.2903/j.efsa.2024.8896

BIOHAZ MANDATE - CPEs IN THE FOOD CHAIN



Carbapenemase-producing E. coli in the food chain in the EU/EFTA

WG Carbapenemases

- **ToR1. State of the art:** BIOHAZ scientific opinion (by March 2025)
- **ToR2. New data generation**
- **ToR3.** State of the art 2027: **BIOHAZ scientific opinion** (June 2027)

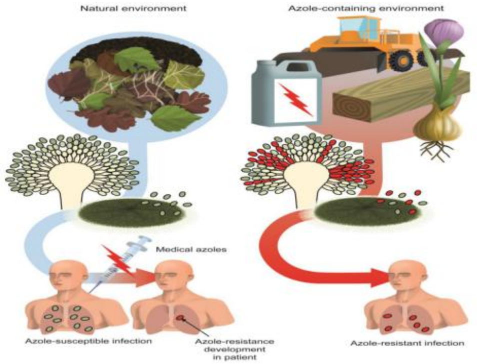
Framework partnership agreement (FPA) "data generation on CPEs in the food-chain"
Contract: 2.5 years (2024-2027)
EURL-AR/NRLs

EC MANDATE - AZOLE RESISTANCE

Use of azole fungicides in the environment, **4 regulatory regimes**:

- Plant protection products (EFSA)
- Biocides (ECHA)
- Industrial chemicals (ECHA),
- e.g. wood preservatives, cosmetics
- Veterinary medicines (EMA)

- Infection in human with *Aspergillus* spp. resistant to treatment with azoles is a health issue:
- Resistance may develop following clinical treatment, use in the environment.



source: *Verweij et al., 2020*

Joint EC Mandate to ECDC-ECHA-EFSA-EEA-EMA(+JRC), overall coordination by EFSA (**BIOHAW+PREV**), different agencies take responsibility to lead different ToRs:

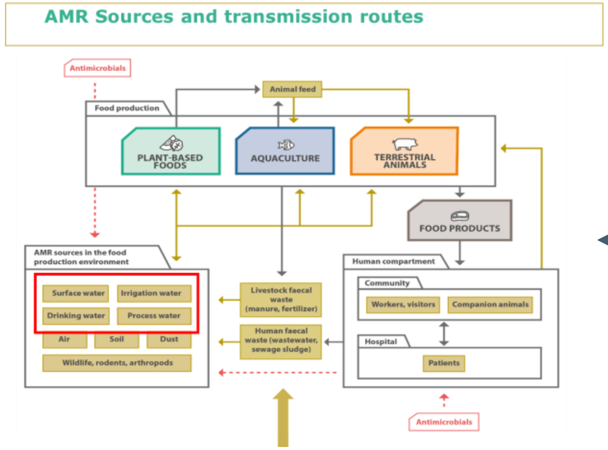
- **Collect data about use** of azole fungicides in all domains other than human medicines
- **Identify causative link** between environmental **use and resistance** development and describe epidemiology
- Assess **risks**
- Identify **risk factors and control options**
- **Identify type of studies to be provided by applicants** for approval of azole substances for different types of use (affecting applications to ECHA, EFSA, EMA)
- Identify data **gaps and research needs**

Deadline: **December 2024**

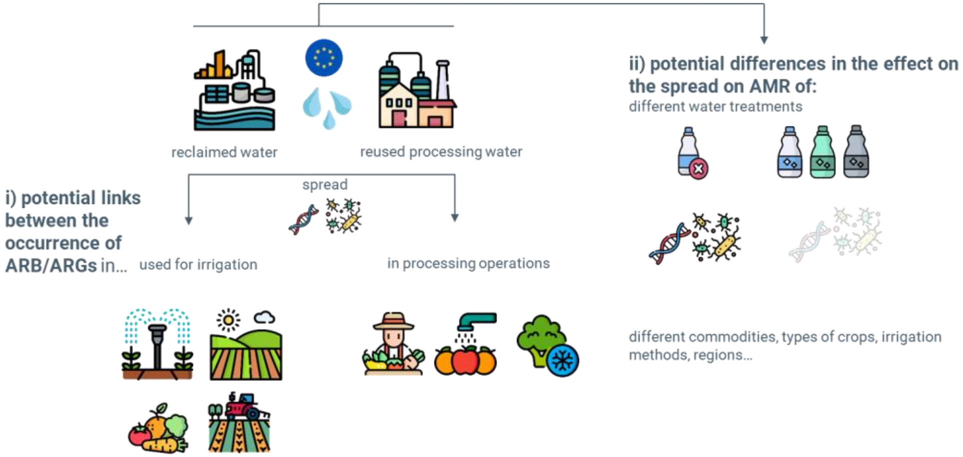
AMR Grants & Procurements

PROCUREMENT: ROLE OF WATER IN THE SPREAD OF AMR-WATERESIST

Direct contract, 950,000 €, …to gain insights on the **occurrence/variety of ARB** and **ARGs**…in order to help **to assess the role of reused water in the spread of ARB and ARGs to fruits/vegetables/ herbs (FVH)** in different European regions



https://efsa.onlinelibrary.wiley.com/doi/epdf/10.2903/j.efsa.2021.6651



reclaimed water

reused processing water

spread

i) potential links between the occurrence of ARB/ARGs in…

used for irrigation

in processing operations

ii) potential differences in the effect on the spread on AMR of:

different water treatments

different commodities, types of crops, irrigation methods, regions…

Timelines: 2024-2027

Objective 1: Optimize ARB/ARGs detection methods: culture, PCR-based, metagenomics

Objective 2: Occurrence ARB/ARGs, using reclaimed water for irrigation

Objective 3: Occurrence ARB/ARGs using “reused/recycled” water during handling/processing

Icons from www.flaticon.com

GRANT – CARBACAMP PROJECT

CarbaCamp
GP/EFSA/BIOHAW/2023/04

September 2023

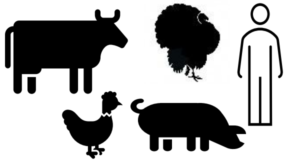
Extension to February 2026 (TBC)

Beneficiary
DTU (Art. 36)

Subcontractor
EUCAST
Development
Laboratories (EDL)

Objectives

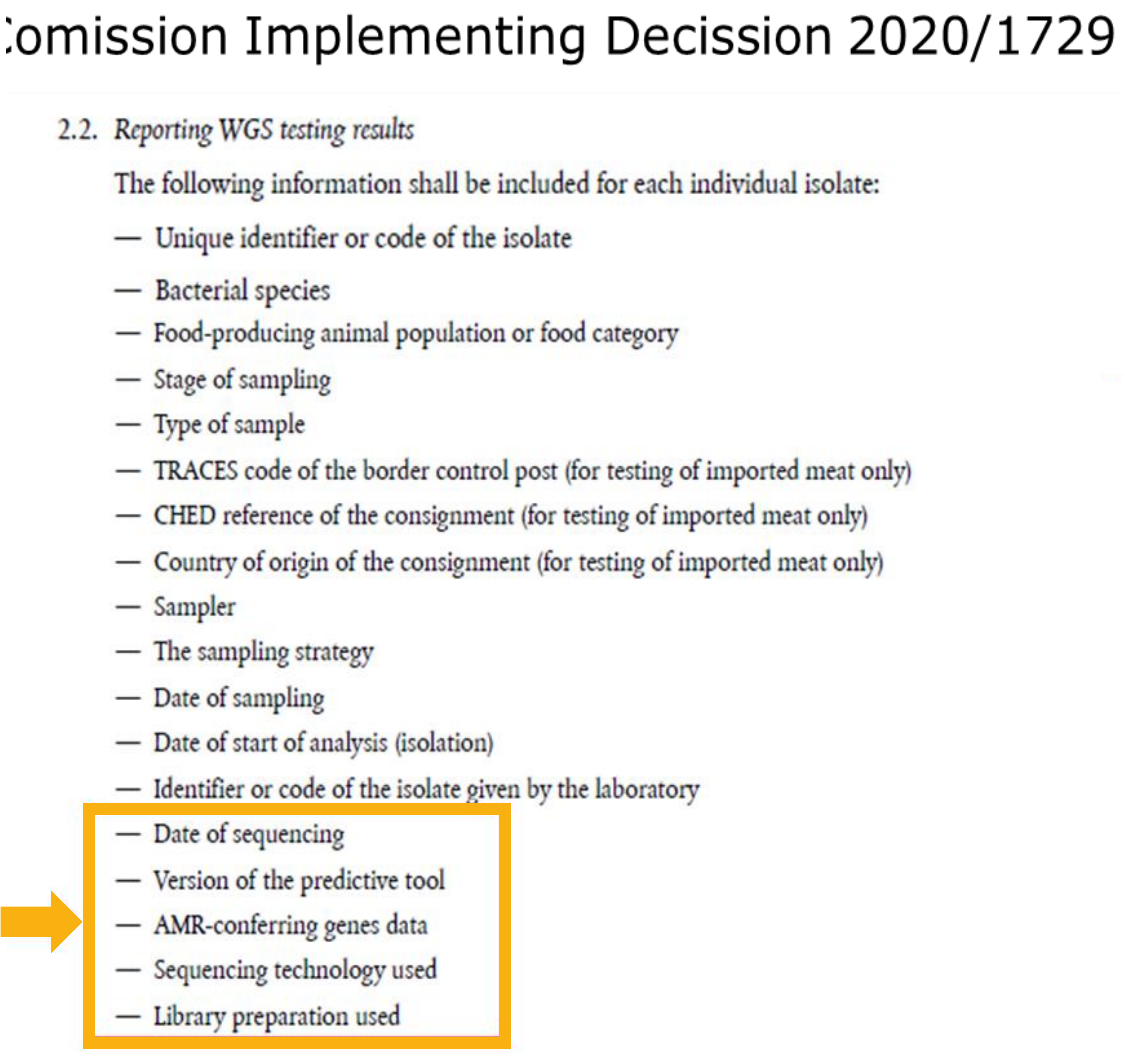
Purpose of the study



- wild-type distribution between *C. jejuni* and *C. coli* in the different species
- ECOFF values ertapenem, imipenem, meropenem
- comparability between the EUCAST and CLSI recommended media for MIC determination of Campylobacter
- monitoring of Campylobacter ? ertapenem imipenem meropenem
- genomic diversity of susceptible and non-susceptible *C. jejuni* and *C. coli*
- Investigating resistance mechanisms

Point for Discussion

AMR GENES – WGS REFERENCE DATABASE

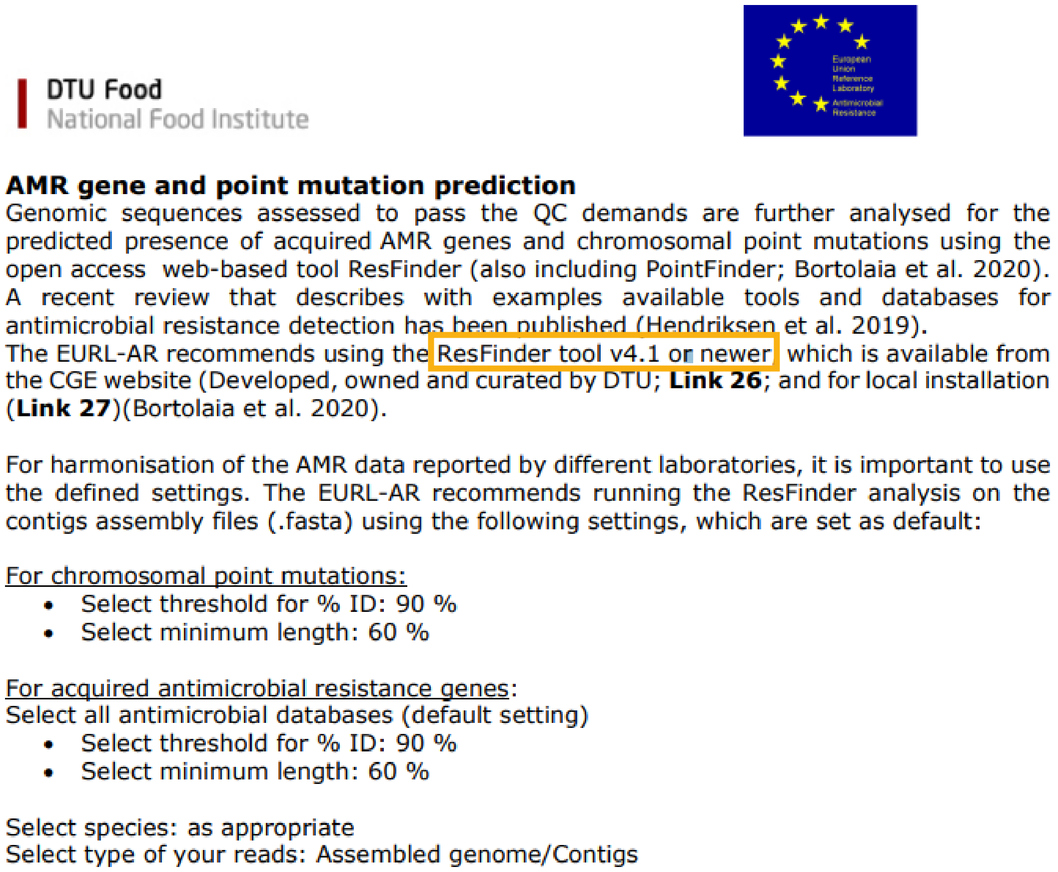


omission Implementing Decission 2020/1729

2.2. *Reporting WGS testing results*

The following information shall be included for each individual isolate:

- — Unique identifier or code of the isolate
- — Bacterial species
- — Food-producing animal population or food category
- — Stage of sampling
- — Type of sample
- — TRACES code of the border control post (for testing of imported meat only)
- — CHED reference of the consignment (for testing of imported meat only)
- — Country of origin of the consignment (for testing of imported meat only)
- — Sampler
- — The sampling strategy
- — Date of sampling
- — Date of start of analysis (isolation)
- — Identifier or code of the isolate given by the laboratory
- — Date of sequencing
- — Version of the predictive tool
- — AMR-conferring genes data
- — Sequencing technology used
- — Library preparation used



DTU Food
National Food Institute

AMR gene and point mutation prediction

Genomic sequences assessed to pass the QC demands are further analysed for the predicted presence of acquired AMR genes and chromosomal point mutations using the open access web-based tool ResFinder (also including PointFinder; Bortolaia et al. 2020). A recent review that describes with examples available tools and databases for antimicrobial resistance detection has been published (Hendriksen et al. 2019). The EURL-AR recommends using the ResFinder tool v4.1 or newer which is available from the CGE website (Developed, owned and curated by DTU; **Link 26**; and for local installation (**Link 27**)(Bortolaia et al. 2020).

For harmonisation of the AMR data reported by different laboratories, it is important to use the defined settings. The EURL-AR recommends running the ResFinder analysis on the contigs assembly files (.fasta) using the following settings, which are set as default:

For chromosomal point mutations:

- Select threshold for % ID: 90 %
- Select minimum length: 60 %

For acquired antimicrobial resistance genes:

Select all antimicrobial databases (default setting)

- Select threshold for % ID: 90 %
- Select minimum length: 60 %

Select species: as appropriate
Select type of your reads: Assembled genome/Contigs

AMR GENES – WGS REFERENCE DATABASE

Which **databases** are you currently using for the detection/ identification of AMR genes?

1. Resfinder/Pointfinder
2. AMRFinderPlus
3. CARD
4. Other databases
5. Combination

Do you use…..

1. Resfinder/Pointfinder alone?
2. Resfinder/Pointfinder in combination with others?

Thanks for your attention!

Acknowledgements

EFSA staff members:

Pierre-Alexandre Beloeil
Mirko Rossi
Ernesto Liebana Criado
Pietro Stella
Beatriz Guerra

STAY CONNECTED

SUBSCRIBE TO
efsa.europa.eu/en/news/newsletters
efsa.europa.eu/en/rss
Careers.efsa.europa.eu – job alerts

LISTEN TO OUR PODCAST
Science on the Menu –Spotify, Apple Podcast and YouTube

FOLLOW US ON TWITTER
@efsa_eu
@plants_efsa
@methods_efsa
@animals_efsa

FOLLOW US ON LINKEDIN
Linkedin.com/company/efsa

FOLLOW US ON INSTAGRAM
@one_healthenv_eu

CONTACT US
efsa.europa.eu/en/contact/askefsa

efsa
EUROPEAN FOOD SAFETY AUTHORITY