

## FINAL REPORT

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### Background

The ECDC Fellowship Programme is a two-year competency-based training with two paths: the field epidemiology path (EPIET) and the public health microbiology path (EUPHEM). After the two-year training, EPIET and EUPHEM graduates are considered experts in applying epidemiological or microbiological methods to provide evidence to guide public health interventions for communicable disease prevention and control.

Both curriculum paths provide training and practical experience using the 'learning by doing' approach at acknowledged training sites across European Union (EU) and European Economic Area (EEA) Member States.

According to Article 9 (6), Article 5 (8) and Article 11a (1) of Regulation (EU) 2022/2370 of the European Parliament and of the Council of 23 November 2022 amending Regulation (EC) No 851/2004 establishing a European centre for disease prevention and control (the ECDC Founding Regulation):

Article 9 (6) 'The Centre shall, as appropriate, support and coordinate training programmes, in particular in relation to epidemiological surveillance, field investigations, preparedness and prevention, response to public health emergencies, public health research and risk communication. Those programmes shall take into consideration the need for training to be kept up-to-date, take into account the training needs of Member States and shall respect the principle of proportionality.'

Article 5 (8) 'By encouraging cooperation between experts and reference laboratories, the Centre shall foster the development of sufficient capacity within the Union for the diagnosis, detection, identification and characterisation of infectious agents that have the potential to pose a threat to public health. The Centre shall maintain and extend such cooperation and support the implementation of quality assurance schemes'.

Article 11a (1) 'The Centre shall establish a EU Health Task Force and ensure that there is a permanent capacity and an enhanced emergency capacity to mobilise and use it. The EU Health Task Force shall provide assistance with regard to requests for prevention, preparedness and response planning, local responses to outbreaks of communicable diseases and after-action reviews in Member States and in third countries, in cooperation with the WHO. The EU Health Task Force shall include the Centre's staff and experts from Member States, fellowship programmes and international and non-profit organisations'.

Moreover, Article 47 of the Lisbon Treaty states that 'Member States shall, within the framework of a joint programme, encourage the exchange of young workers.' Therefore, ECDC initiated the two-year EUPHEM training programme in 2008. EUPHEM is closely linked to the European Programme for Intervention Epidemiology Training (EPIET). Both EUPHEM and EPIET are considered 'specialist pathways' of the two-year ECDC fellowship programme for applied disease prevention and control.

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This final report describes the output of the fellow and the competencies they acquired by working on various projects, activities, theoretical fellowship training modules, other modules or trainings and international assignments or exchanges during the fellowship.

## Pre-fellowship short biography

**Vera Mónica Martins Gonçalves Manageiro** has a PhD in Biology (2011), speciality in Microbiology and a BSc. Degree in Applied Chemistry – Biotechnology. She has been a researcher at the National Reference Laboratory of Antibiotic Resistances and Healthcare-Associated Infections, Department of Infectious Diseases, at the National Institute of Health Doutor Ricardo Jorge. Vera has gained expertise and skills in the field of pathogen genomics by enrolling in a Postgraduation in Bioinformatics and Computational Biology as well as several courses. She has also developed teaching activities as assistant professor in General Microbiology, Molecular Microbiology, Food Microbiology and Bioinformatics. Vera's research focused on antibiotic resistance, particularly exploring how resistome, virulome, and mobilome connect various reservoirs from a One Health perspective. Since 2010, she has also been a European Centre for Disease Prevention and Control operational contact point for Antimicrobial Resistance/European Antimicrobial Resistance Surveillance Network (EARS-Net) in Portugal. Vera's initial goals for the EUPHEM fellowship were to expand her knowledge in public health microbiology and epidemiology.

## Results

The objectives of these core competency domains were achieved partly through project and activity work and partly by participating in the training modules. Results are presented in accordance with the EUPHEM core competencies, as set out in the ECDC Fellowship Manual<sup>1</sup>.

### 1. Epidemiological investigations

#### 1.1. Outbreak investigations

##### *Ongoing monkeypox virus outbreak, Portugal, April-May 2022*

Supervisor: Jorge Machado

Category: Hepatitis B and STDs

On 3 May 2022, five males with atypical skin ulcerative lesions with similar body distribution presented at Centro Hospitalar Universitário de Lisboa Central sexually transmitted infection (STI) outpatient clinic and at the CheckpointLX, a community based STI clinic targeted at men who have sex with men (MSM). On 16 May, following an EpiPulse alert from the UK of a confirmed case for mpox in an individual with similar genital lesions, an initial virological screening PCR assay for orthopoxvirus was carried out and an emergency management team was assembled to follow the alert. On 17 May, the National Institute of Health Doutor Ricardo Jorge provided laboratory confirmation of the first three cases. As of 27 May, 96 cases of mpox were confirmed in Portugal. In this study, we collected in depth information on a subset of 27 cases. We obtained information on demographic characteristics, clinical presentation, and exposure from all cases through face-to-face and phone interviews, using standardised case-investigation forms. The median age was 33 years (range 23-61), and the patients were all male. The earliest symptom onset date was 29 April. Almost all cases (25, 93%) live in the Lisbon and Tagus Valley health region. A third of the cases (nine, 33%) had attended a sex venue in Portugal, while five (18%) reported having travelled abroad. Preliminary genetic data suggest importation from West Africa. Although monkeypox virus was classified as moderately transmissible, studies have described an increasing genetic adaptation to a human host, with enhanced potential for human-to-human transmission. Public health authorities engaged with the LGBTIQI community, particularly community leaders, early on, on risk communication and social mobilisation of the community to ensure that information on the infection signs and symptoms and how to reduce transmission was promptly shared.

Role: Co-investigator. The fellow attended the follow-up meetings, contributed with data analysis, and co-wrote the draft of the Rapid Communication with the preliminary results of the outbreak investigation (Publications, manuscript section 7.1.1, no. 1).

<sup>1</sup> European Centre for Disease Prevention and Control. European public health training programme. Stockholm: ECDC; 2020. Available from: <https://www.ecdc.europa.eu/en/publications-data/ecdc-fellowship-programme-manual-cohort-2021>

## ***Epidemiological investigation of a Norovirus GII.4 Sydney variant outbreak in a long-term care facility in Portugal, 2022***

Supervisors: Nuno Rodrigues and Rita de Sousa

Category: Food and waterborne diseases

An outbreak of norovirus (NoV) occurred among 60 residents and 48 staff members of a long-term care facility between the 25 April and 7 May 2023. A total of 43 people met the probable case definition (diarrhoea, vomiting, or nausea between 25 April and 14 May 2022), and eight were laboratory-confirmed. The overall attack rate was 41.4% (45.5% among staff and 38.3% among residents). The highest attack rates were observed in male residents (50.0%), and in staff who participated in personal hygiene tasks (50.0%). Out of 11 symptomatic individuals of whom stool samples were tested, six residents and two staff members were positive for NoV GII, identified further as NoV GII.4 Sydney [P16] variant by genotyping. NoV was also found in two of the six surface swabs taken from the toilet faucet of residents' living rooms, whereas all the 15 food samples collected from the kitchen were negative. A lack of soap and alcohol-based hand rubs in the shared toilets was reported. Although the source of NoV introduction in the long-term care facility (LTCF) could not be identified, virus spread was likely facilitated by deficiencies in the hygiene conditions. No more cases were detected after the 7 May and the outbreak was considered over on the 14 May. The outbreak was likely controlled by ensuring that proper disinfection procedures were in place and guidelines for the management of norovirus outbreaks were followed.

Additional microbiological analysis was conducted to assess the persistence and dissemination of the NoV GII.4 Sydney [P16] variant in the region. The genetic analysis of several NoV clusters that occurred in the same region was integrated into this investigation. A scientific article is planned to present the results obtained in this additional study (submission after the end of the fellowship).

Role: Co-investigator. The fellow participated in all phases of the investigation: 1) Field investigation at the LTCF, where she contributed with the geomapping of the cases epidemiological data collection (face-to-face and telephone surveys), line listing, and implementation of control measures; 2) conducted the microbiological analysis of stool specimens, virus characterisation by genotyping, epidemiological and microbiological data analysis; and 3) outbreak report co-writing (publications, other reports section 7.1.2, no. 1). Additionally, the fellow was co-author of two abstracts submitted to international conferences (Conference presentations, section 7.2, nos. 1 and 2). For the additional investigation, the fellow participated in the microbiological and phylogenetic analysis of the NoV clusters/outbreaks that occurred in the same region and will contribute to the planned manuscript (Publications, manuscript section 7.1.1, no. 2).

## ***Challenges of investigating a large-scale outbreak of norovirus gastroenteritis in a hotel, Algarve, Portugal 2022***

Supervisor: Rita de Sousa

Category: Food and waterborne diseases

Norovirus (NoV) is a leading cause of gastroenteritis outbreaks worldwide. There is no surveillance network to monitor NoV activity and outbreaks in Portugal, despite NoV being a significant cause of morbidity and cost within healthcare systems. This study presented an epidemiological investigation of a NoV gastroenteritis outbreak that occurred in a hotel in Portugal in August 2022. On 19 August 2022, several cases of acute gastroenteritis were identified in guests and staff of a hotel in Albufeira, Algarve, with onset of symptoms in the afternoon of 18 August 2022. There were 244 cases identified up to 26 August 2022, primarily involving Portuguese families with parents aged 40-50 and children aged 0-19. Five cases were hotel employees. Common symptoms included vomiting, nausea, abdominal pain, and diarrhoea. The samples were sent to a local microbiology laboratory. According to the laboratory investigation, NoV was detected in stool samples from eight probable cases. The health authorities took immediate action to reinforce hygiene and disinfection measures. A meeting was held with the hotel management to review contingency plans for probable future outbreaks. At the National Institute of Health Doutor Ricardo Jorge, NoV GI.3 was detected in stool specimens, while food samples tested negative for NoV and other commonly implicated microorganisms. No other NoV genotypes were detected so GI.3 genotype was suspected of being the main cause of the outbreak. The most likely hypothesis is that the source of the outbreak was in the hotel's common areas, with subsequent person-to-person transmission occurring primarily within one of the buildings. Although the exact source was not identified, this investigation underscores the need for improvement in outbreak prevention and control measures, including the development of a plan, the establishment of an outbreak response team, and the enhancement of regional laboratory capacity.

Role: Co-investigator. The fellow participated in the microbiological investigation, including the phylogenomic analysis, and helped to write and review the manuscript (Publications, manuscript section 7.1.1, no. 3). The fellow also co-wrote an abstract, that was submitted to 16th European Public Health Conference (EPHC) 2023 but was not accepted (Conference presentations, section 7.2, no. 3).

## **Outbreak of carbapenemase-producing *Enterobacterales* in Braga, Portugal, 2022**

Supervisor: Manuela Caniça

Category: Healthcare-associated infections and antibiotic resistance

Since March 2022, an increase in the rate of carbapenemase-producing *Enterobacterales* (CPE) was noticed by the microbiology laboratory. Preliminary epidemiological and microbiological studies confirmed the existence of an outbreak. A multi-disciplinary team was organised to investigate and control it. In this study we aimed to determine the source of the outbreak, identify the risk and exposed people; and to understand the mode of CPE transmission. A multi-disciplinary investigation was conducted including descriptive, analytical, and molecular epidemiology, environmental screening, and assessment of infection prevention and control measures. CPE screening was performed by molecular methods. Whole-genome sequencing and subsequent phylogenetic analysis was used to characterise the genetic relationships of 74 (59 clinical and 15 environmental) isolates collected through 2022. We further investigated the risk factors associated with the emergence of new cases in non-CPE wards (cohorts of CPE-negative admitted patients), by conducting a prospective observational cohort study using a structured questionnaire. Patients aged  $\geq 75$  years (RR=3.3; 95%CI:1.2-9.0), hygiene and/or feeding care dependent (RR=2.7; 95%CI:1.3-5.7), and/or submitted to secretion aspiration (RR=2.6; 95%CI:1.4-4.9) were considered risk factors for the emergence of cases in non-CPE wards. The microbiological results showed the existence of multispecies KPC-, IMP-, and/or NDM-producing isolates. Genetically indistinguishable clinical and environmental (e.g. shower drains, room sink, cleaning service cart) isolates were found on the same room/ward, with epidemiological links. The KPC-3-producing *Klebsiella pneumoniae* ST45 clone was the main clonal lineage responsible for the ongoing outbreak. Overall, the epidemiological and laboratory results suggested that the hospital environment, with possible transmission during patient handling in hygiene care and/or feeding, was playing a key role in the spatial and temporal CPE dissemination in the hospital. The control measures already in place were reinforced, specifically in-depth disinfection of hospital surfaces, adherence to hygiene procedures among staff and visitors, and indoor air quality monitoring. To date, the outbreak is contained, and CPE has become endemic in the hospital with a low frequency of isolation (1-2% of incidence vs 8-10% during the outbreak).

Role: Co-investigator. The fellow was involved in all the 10-steps of the outbreak investigation. This included site visits to discussions on control and prevention measures, clinical and environmental microbiology analyses, and further investigations if needed. The fellow played a role in questionnaire and epidemiological study design, conducted microbiological investigations, and analysed epidemiological and microbiological information, including WGS data. Additionally, the fellow contributed to results communications: co-drafted the internal outbreak reports (in Portuguese; Publications, other reports section 7.1.2, no. 2) and is in the process to submit a manuscript to a peer-reviewed journal (Publications, manuscript section 7.1.1, no. 4).

### **Educational outcome**

Participation in the different outbreak investigations allowed the fellow to apply concepts of microbiology and epidemiology in outbreak situations. The fellow developed knowledge and skills in several aspects of outbreak epidemiology (the ten steps of an outbreak investigation, including analytical studies, data management and STATA analysis), actively participated and engaged with multidisciplinary outbreak teams, performed laboratory typing methods, and wrote outbreak reports. The fellow was also able to deepen her skills in microbiology and communication in outbreak situations.

## **1.2. Surveillance**

### **Tracking epidemiological shifts in hepatitis A in Portugal: a comparison of seroprevalence in 2001-2002 and 2015-2016**

Supervisor: Rita de Sousa and Baltazar Nunes

The global incidence of hepatitis A virus (HAV) infection decreased with improvements in sanitation/hygienic conditions, in some regions combined with childhood vaccination. Likewise, in Portugal, regional and/or group-specific studies have shown over time a decrease in the number of cases and consequently in the prevalence of antibodies to HAV in the population. Our aim was to estimate HAV seroprevalence and compare population-level immunity over time, based on the two nationwide serosurveys. We analysed data from two cross-sectional population-based serosurveillance studies performed in 2001–2002 and 2015–2016 in the Portuguese population aged  $\geq$ two years. We estimated seroprevalence as the proportion of individuals with detected anti-HAV IgG antibodies in serum samples weighted for the population distribution by age, sex and district and stratified by population characteristics. Associations between socio-demographic and prevalence was measured with adjusted prevalence ratios (PR) and corresponding 95%CI. Overall seroprevalence of anti-HAV was 67.3% (95%CI:64.2%-70.3%) in 2001–2002 (n=1 642) compared to 56.3% (95%CI:52.4%-60.2%) in 2015–2016 (n=2 692), showing an absolute decrease of 11% between studies. There were no significant differences between sex, residence district or nationality. Regarding birth cohort analysis, people not immune in 2001–2002 remained susceptible in 2015–2016, suggesting that having a higher seroprevalence is more related with the birth cohort (cohort effect) than with a particular point in time. For instance, in the birth cohort 1981–1990 the seroprevalence was 16.7% (95%CI:12.4–22.1) in 2001–2002 and 18.7% (95%CI:12.3–27.3) in 2015–2016, with a non-significant increase of 2%. Within each survey, the seroprevalence was higher for people with lower educational level. Between surveys, the PR ranged from 1.0 (95%CI:1.0–1.1) for no formal instruction/basic to 1.3 (95%CI:1.1–1.6) for secondary/higher level of education.

This study shows a trend of increasing age of the susceptible population, reflecting the improved living conditions in last decades. After April 1974, there was a significant political investment in the improvement of water, sanitation, and hygiene conditions which could explain the switch in the HAV immunity for generations born after the 1980s observed in this study. Considering the age of the population immunised to HAV had shifted towards older age groups, these results could be of great value in the future revision of the HAV vaccination strategies in Portugal.

Role: The fellow carried out the literature review, data analysis and interpretation of laboratory surveillance data, including data cleaning and recoding, databases uniformisation and aggregation, and statistical analysis using STATA. The fellow was involved in draft and submission of abstracts for international conferences and meetings (Conference presentations, section 7.2, no. 4 and 5; Other presentations, section 7.3, no. 1) and of a manuscript to a peer-reviewed journal (Publications, manuscript section 7.1.1, no. 5).

### **Trends in antibiotic resistance in Portugal: analysis of surveillance data from 2015 to 2020**

We are seeing, an increasing number of patients with multidrug-resistant bacteria infection in Portugal and across Europe. In this study, we aimed to analyse antibiotic resistance (AR) surveillance data collected by Portuguese hospitals to describe the current situation and evaluate the need for improved public health strategies. The surveillance data used in this study was collected from Portuguese laboratories reporting AR data from invasive isolates from 2015 to 2020 (national population coverage: 97%). Susceptibilities were determined with methods in use by the reporting laboratories and evaluated by an external quality control program. The statistical significance of trends was calculated based on data from the last six years (2015–2020) and assessed by a chi-square test for trend. A value of  $p < 0.05$  was considered significant. The occurrence of methicillin-resistant *Staphylococcus aureus* (MRSA) in Portugal significantly decreased from 47%/2015 to 30%/2020 ( $p \leq 0.001$ ). Regarding *Streptococcus pneumoniae*, Portugal showed an increasing penicillin-non-wild-type trend (11%/2015 to 14%/2020,  $p = 0.349$ ). *Enterococcus* spp. displayed a decreasing trend for vancomycin-resistant isolates (*E. faecium*: 20.2%/2015 to 7.6%/2020; *E. faecalis*: 2.0%/2015 to 0.5%/2020,  $p \leq 0.001$ ). Carbapenem- and colistin-resistant *Escherichia coli* remained sporadic in 2020 (0.3% and 0.6%, respectively). The occurrence of carbapenem-resistant *Klebsiella pneumoniae* indicated an increasing trend (4%/2015 to 14%/2020,  $p \leq 0.001$ ). Indeed, for *K. pneumoniae*, a statistically significant increase was observed for all antimicrobial groups under surveillance except for colistin ( $p = 0.539$ ). The trends for colistin-resistant *Pseudomonas aeruginosa* and *Acinetobacter* spp. increased significantly during the study period ( $p \leq 0.001$  and  $p = 0.002$ , respectively). The AR situation in Portugal displays wide variations depending on the bacterial species and antibiotic group. The decreasing frequency of invasive-MRSA in the recent years it is an important achievement. However, strategies to minimise further spread of carbapenem- and colistin-resistant isolates are urgently needed. These may involve strict infection control, antimicrobial stewardship programs, active surveillance, rapid diagnostic tests, and promoting proper hand hygiene.

Role: This surveillance activity was performed by the fellow within her role as ECDC operational contact point for EARS-Net in Portugal. The fellow was involved in the process of laboratory surveillance data collection at national level and respective data analysis, interpretation of the epidemiological and microbiological data (current trends), and statistical analysis using STATA. The fellow drafted and submitted an abstract, accepted as oral poster presentation in international conference and meetings (Conference presentations, section 7.2, no. 6; Other presentations, section 7.3, no. 2).

### **Epidemiological Surveillance at mass gathering events (Andanças and BOOM Music/Dance Festivals)**

Supervisor: Ricardo Mexia

Mass gatherings are events that bring together a large number of people in one place for a specific purpose and a defined period of time, which can put local systems under strain. In the case of music festivals, health risks are increased due to high concentrations of participants, temporary catering facilities, high alcohol consumption, and recreational drug use. Therefore, it is important to quickly detect any situation that requires intervention to protect the health of participants and the community. Andanças is a festival that promotes popular music and dance. The 2022 edition took place in Campinho, Portugal, with around 1 500 participants. The Boom Festival is a biennial, multidisciplinary, and psychedelic festival that takes place in Idanha-a-Nova, Portugal. The 2023 edition was held from 20–27 July, with 39 452 participants. Both festivals had an epidemiological surveillance team (EpiTeam) recording the healthcare incidents. In Andanças, 185 incidents were documented, while the Boom Festival registered 2 527 until the 26 July, as part of active and syndromic surveillance. Globally, the most common complaints included blisters/wounds and osteoarticular diseases. Both festivals had cases of gastroenteritis, but no clear epidemiological link could be found amongst each festival attendees. In Andanças, the EpiTeam was notified about a suspected case of measles with an initial IgM-positive test. The participant provided blood and urine samples for confirmation, which were sent to the National Reference Laboratory for vaccine-preventable diseases, INSA. The case was not confirmed by molecular biology techniques. In BOOM, the EpiTeam managed to obtain an intact tick from a male who'd been bitten. The tick was stored and sent to the Center for the Study of Vectors and Infectious Diseases (CEVDI/INSA) for investigation. It was identified as an adult female *Hyalomma lusitanicum*. No tick-borne pathogens were detected (molecular methods) in the investigation. In conclusion, the surveillance measures implemented were effective in identifying and managing potential health risks.

This highlights the importance of proactive surveillance and timely interventions in ensuring the safety and well-being of attendees at mass gathering events.

**Role:** The fellow was integrated with the EpiTeam responsible for the early detection and response of Public Health Threats at Andanças 2022 and BOOM 2023 Festivals. The fellow handle real-time data collection within shifts, covering 24h, at the different Health Care facilities ("Espaço Saúde" at Andanças; BOOM Medical Service and Kosmicare at BOOM). The fellow also participated at the daily briefing with Authorities and Production team at BOOM. Additionally, she provided microbiological assistance, such as collection and transporting specimens for microbiological investigation.

### **Educational outcome**

The fellow acquired substantial experience in several surveillance systems. The fellow carried out data collection and analysis of sentinel laboratory-based surveillance systems and collaborated in active and syndromic surveillance for two mass gathering events. The fellow also gained insight into the significance of seroprevalence studies in monitoring infectious diseases, contributing to the formulation of public health interventions. Furthermore, participating in the EpiTeam during mass gathering events exposed the fellow to epidemiological fieldwork under demanding circumstances, such as extreme temperatures and unconventional working hours. This valuable opportunity allowed the fellow to assess her skills, resilience, and ability to adapt to diverse scenarios.

## **2. Applied public health microbiology and laboratory investigations**

### **Evolutionary study of the genetic basis of *Legionella pneumophila* ST1905 in Portugal, 2014–2022**

Supervisor: *Paulo Gonçalves*

The second largest-to-date outbreak of Legionnaires' Disease (LD) occurred in Lisboa, Portugal, in 2014, associated with the ST1905 genotype of *Legionella pneumophila* subspecies *fraseri* serogroup 1. Since 2014 this strain has been sporadically identified in the region in the context of clinical and related environmental investigations. The aim of this study was to explore the 2014–2022 molecular evolution and spatiotemporal dynamics of twelve ST1905 strains, isolated between 2014 and 2022. Illumina's Nextera XT and MiSeq were used for DNA library preparation and sequencing. Genomes were *de novo* assembled using the INNUca pipeline. Snippy was used for reference-based mapping and SNP/indel analysis, using PtVFX/2014 as reference. ReporTree identified core genome SNPs, used for creating a maximum likelihood phylogenetic tree with MEGA. All ST1905 clinical and environmental isolates analysed were genetically similar to PtVFX/2014, with the observed microevolution being marked by ten SNPs, four indels and one recombination event. All strains presented a recombination event in the T4ASS region. Two deletions were detected in one strain, on a region corresponding to a genomic island harbouring a *lvh/lvr* T4ASS cluster and on the CRISPR-associated intergenic region. In-depth phylogenetic analysis showed that the isolates did not cluster by year of isolation, and both clinical and environmental isolates associated to a particular location clustered apart, supporting the persistence of the strain in that location and the linkage between different events. Our study suggests that ST1905 has been conserved over time and seems well adapted and potentially persistent in diverse and geographically dispersed environments. The intrinsic pathogenic and virulence characteristics of this strain and the potential for spreading to other locations not previously affected by the 2014 outbreak may result in serious implications to public health, highlighting the importance for continuous monitoring and control.

**Role:** The fellow reviewed the literature and made a study protocol for the analysis. The fellow was involved in the analysis and interpretation of epidemiological and comparative genome analysis including phylogenetic and gene variation investigation. The fellow communicated the study results as a manuscript in a peer-reviewed journal (Publications, manuscript section 7.1.1, no. 6) and an oral presentation at a national conference (Conference presentations, section 7.2, no. 7). The presentation was awarded with Best Oral Communication Prize.

### **Genomic profiles of *Rickettsia conorii* strains isolated from patients with Mediterranean spotted fever, 1994-2022, Portugal**

Supervisor: *Rita de Sousa*

*Rickettsia conorii* is a Gram negative and obligate intracellular pathogen that causes Mediterranean spotted fever (MSF), a serious disease and well-known rickettsiosis in the Mediterranean basin. In Portugal, MSF is the most important tick-borne disease of public health concern and has one of the highest incidence rates, when it is compared with other endemic countries in the Mediterranean basin. The disease is caused by two strains: *R. conorii* Malish and *R. conorii* Israeli spotted fever (ISF), both transmitted by the brown dog tick *Rhipicephalus sanguineus*. A large clinical prospective study including 140 Portuguese patients, with confirmed molecular identification of the causative rickettsial strain, documented that ISF strain was more virulent than Malish strain, with more significant gastrointestinal symptoms and more altered hepatic parameters. Moreover, patients infected with ISF had a 2.81 higher risk of death than patients infected with Malish strain. The aim in this study was to perform a comparative intraspecies genomic analysis of *R. conorii* strains using NGS as a new tool to get more deep insights of potential *R. conorii* strain differences that were not identified with isolates partial sequences genes (*ompA*, *ompB*, *gltA*) obtained in the previous study.

For this purpose, we sub-cultured more than 100 isolates from Portuguese patients admitted to eight hospitals with a clinical diagnosis of MSF during the last 27 years (1994–2022). The methodology included the following steps: 1) sub-culture of *Rickettsia* isolates and DNA extraction using a specific protocol developed for this purpose; 2) selection of other type of *Rickettsia* positive specimens (e.g., blood, skin biopsies); 3) whole-genome sequencing (WGS, with and without target enrichment) and assembly; 4) comparative genome analysis; 5) analysis of anonymised epidemiological and clinical data. We were successful to sequence 70 *R. conorii* strains (31 *R. conorii* Malish and 39 *R. conorii* ISF) from culturable isolates plus 24 obtained directly from PCR positive clinical specimens. Phylogeny and global polymorphism analysis (parSNP), clustering; fine-tune analysis of the inter- and intra-subspecies diversity and analysis of the major virulence factors in *Rickettsia* genome (e.g., rOmpA/Sca0, rOmpB/Sca5, Sca1, RickA) are in progress.

Role: The fellow contributed to the isolation of *Rickettsia*, an obligatory intracellular bacteria, from patients samples and to the optimisation of the time-consuming DNA extraction protocols for next-generation sequencing (NGS), including the targeted whole-genome capture and sequencing directly from non-culturable samples. The fellow also analysed the genomic data (genomic analysis of the inter- and intra-subspecies diversity) and will contribute to the preparation of the manuscript to be submitted after the end of the fellowship (Publications, manuscript section 7.1.1, no. 7). The fellow communicated the study results as an oral presentation (Other presentations, section 7.3, no. 3).

### **Genomic surveillance of SARS-CoV-2 (COVID-19) in Portugal**

Since the beginning of the COVID-19 pandemic, the National Institute of Health Doutor Ricardo Jorge (INSA) has been acting as National Reference Laboratory for SARS-CoV-2, through both the National Reference Laboratory for Influenza and other Respiratory Viruses, and the Bioinformatics Unit. INSA settled a genome-based molecular surveillance network for SARS-CoV-2 in Portugal, in collaboration with a nationwide network of hospitals/labs, who on a weekly base send SARS-CoV-2 positive samples (either clinical specimens or RNA) for genomic analysis. The aim of this activity was to understand the laboratory and bioinformatics workflow of the ongoing SARS-CoV-2 genomic surveillance in Portugal, from the SARS-CoV-2 positive samples reception to the production of the INSA weekly 'situation' reports with major highlights released periodically to participating laboratories, national and regional public health authorities, and other stakeholders. Specific activities conducted included 1) RNA samples reception and pre-sequencing protocol for SARS-CoV-2; 2) Illumina Library Preparation and NGS; 3) Bioinformatics analysis using INSAFLU online platform; and 4) Preparation of the weekly report. The weekly reports were then provided to public health authorities and the general-public, through a specific website (<https://insaflu.insa.pt/covid19>) which allowed the analysis of the SARS-CoV-2 genetic diversity and geotemporal dynamics, based on state-of-the-art methodologies for real-time tracking pathogen evolution tools.

Role: The fellow was involved in all stages of the workflow that takes place after RNA extraction up to the weekly report, as an observer, and summarised her experience in a report.

### **Educational outcome**

The fellow expanded her microbiological knowledge in virology, bacteriology, and public health. The fellow was familiarised with all stages of conducting a public health research project, from identification of the public health problem, reviewing literature, writing study protocols, understanding laboratory methods, analysing data, and writing scientific manuscripts. Vera understood the use and limitation of diagnostic and typing methods and their interpretation. The fellow also improved her expertise in analysis of genomic sequence data and in the diversity of methodologies used in the rapid response to an important public health problem.

## **3. Biorisk management**

### **COVID-19 response team for molecular laboratory diagnosis at INSA**

The COVID-19 pandemic, caused by the SARS-CoV-2 virus, was officially declared by the World Health Organization (WHO) on 11 March 2020. Laboratory diagnosis was crucial in identifying both symptomatic and, more importantly, asymptomatic cases. This plays a vital role in implementing strategic measures to control and disrupt the transmission chains of the virus. At the National Institute of Health Doutor Ricardo Jorge (INSA), non-essential laboratory activities were suspended. Since the beginning of the COVID-19 pandemic, INSA acted as National Reference Laboratory for SARS-CoV-2, through the National Reference Laboratory for Influenza and other Respiratory Viruses. Specialised teams were established to contribute to the effective identification and containment of the virus through reliable and efficient testing, ultimately helping to control the spread of SARS-CoV-2 in the community. At INSA, the workflow for a COVID-19 diagnostic test involved several steps. Samples, such as nasal swabs or saliva, were collected by healthcare professionals from individuals suspected of having COVID-19 or for screening purposes. These samples were labelled and transported to INSA. After the samples arrived at INSA, the following tasks were performed: 1) sample reception; 2) sample preparation; 3) SARS-CoV-2 detection and analysis; and 4) result reporting and delivery. As of May 5, 2023, the WHO has lifted the Public Health Emergency of International Concern (PHEIC) for COVID-19. However, the pandemic is not over yet, and COVID-19 is still considered a pandemic, and monitoring the impact of COVID-19 and the effectiveness of prevention and control strategies remains a public health priority.

Role: Since the beginning of the COVID-19 pandemic, the fellow was involved in the molecular laboratory diagnosis as a member of the COVID-19 response teams at INSA, being team leader from October 2020. The fellow performed all stages of the process, from receiving and preparing the samples in a BLS3 environment, to reporting and delivering the results. As a team leader, the fellow was entrusted with the additional task of effectively coordinate and manage the activities of the team to ensure accurate and timely testing for the detection of the SARS-CoV-2 virus. The fellow summarised her experiences and lessons learned in a final report.

### ***Risk assessment as part of the Biorisk and Quality Management module***

As part of the Biorisk and Quality Management module, the fellow was asked to use the BioRAM Lite form to conduct biosafety and biosecurity risk assessment for a laboratory culturing Multi-Drug-Resistant Titan Blue (MDR-TB) for antibiotic susceptibility testing (scenario). The tool provides a process to identify and prioritise biorisk systematically and logically including a set of questions which are scored using ordinal values and the mathematical equations to combine the answers into likelihood (LOI) and consequence of disease (COD) values. The models consider factors such as the properties of the biological agents (e.g., infectious dose, incubation period, ability to mutate in host or environment), the lab security practices (e.g., available equipment), in-place mitigation measures (e.g., immunisation, post infection treatment), the routes of infection in humans and animals, and the likelihood of local malicious activity in relation to the agents. In the given scenario, the risk of direct exposure by inhalation to individuals in the laboratory was found to be high (LOI=3.25/COD=2.21), while the same risk was moderate to the human and animals in the community (human: LOI=2.79/COD=2.32; animal: LOI=2.79/COD=3.71). The risk by ingestion was also moderate for individuals in the laboratory (LOI=2.25/COD=2.21).

### ***Educational outcome***

The fellow has prior experience in biorisk management, having worked in BLS2 and BLS3 and completed relevant courses in the field ('Biosafety and Biosecurity Training Course at BSL2 and BSL3'; 'Transport of Infectious Diseases Substances'). Nevertheless, the fellow's activities presented above increased her understanding of applying biorisk management concepts, biosafety and security risk assessments, and mitigation procedures.

## **4. Quality management**

### ***National Reference Laboratory (NRL) External Audits: NRL for Legionella, NRL for Salmonella, and NRL for Antibiotic Resistance/Healthcare-Associated Infections***

In October 2021, at the Department of Infectious Diseases, at INSA, the fellow participated in three external audits, two as an observer (URGI: National Reference Laboratory for Gastrointestinal Infections - *Salmonella* and URIR: National Reference Laboratory for Respiratory Infections – *Legionella* and *Neisseria meningitidis*) and one as an auditee (URRA: National Reference Laboratory for Antibiotic Resistance and Healthcare-Associated Infections). The audits covered the applicable technical and management requirements of the normative references NP EN ISO/IEC 17025:2018 (General requirements for the competence of testing and calibration laboratories) and NP EN ISO 15189:2014 (Medical laboratories — Requirements for quality and competence), to verify conformity and its implementation in the quality management system. The audits were performed according to the Accreditation Technical Annex E0014-1, and included processes management, quality control, technical references, documentation, storage of reagents, data analysis and communication of results. This involved verifying all procedures from the time the specimen arrives at the laboratory until the results are sent to the health unit which requested them, and the specimen is stored in the biobank. In general, the technical competence of employees responsible for carrying out accredited tests was evidenced, as well as knowledge of the management system procedures relevant to the performance of their activities. All laboratories generally complied with the criteria, including the evaluation of the laboratory performance for the specific tests by EQAs, and internal quality controls. A minor non-conformity was found in subsection 5.1 (Technical requirements: personnel) of the ISO15189 in URGI. Corrective actions were put in place for the non-conformity. No corrective measures were necessary for URIR lab under the audited methods. In external audit at URRA, no non-conformities or improvement opportunities were identified, demonstrating that the lab is working under the quality standards and no corrective measures were necessary. The fellow summarised the results and learning outcomes in a final report.

### ***Quality management module homework: laboratory audit***

On the 19 May 2022, the fellow performed an audit on the process management, quality control and the documentation available at INSA's laboratory. The laboratory is accredited by the Portuguese Institute of Quality (IPQ), which is the national organisation that manages and promotes the development of the Portuguese System for Quality (SPQ). This short assessment confirmed that both the 'Process Management and Quality Control' as well as 'Documentation' are very well registered in general. The laboratory has departments to perform specific tasks, namely receive, register, and distribute internal and external samples (ex. from hospitals; external laboratories; etc.), perform routine and specific clinical analysis, and maintain a central warehouse. Few non-conformities or improvement opportunities were identified, demonstrating that the laboratory is working under the quality standards. One of the few indicators that did not achieve 100% was a lack of process for reviewing client feedback, analysing any errors, and reporting to laboratory personnel and top management; and the lack of surveillance and outbreak response documentation. The overall quality of operational and facility management was high and no major deviations affecting biosafety were identified at the facility.



### ***Educational outcome***

The fellow was already familiar with audits and quality systems, being the Deputy Quality Manager at the NRL for Antibiotic Resistance and Healthcare-Associated Infections since 2018. The fellow consolidated the previous experience with quality control of phenotypic antibiotic susceptibility tests, being able to demonstrate with the remaining team all processes and records requested by the audit team. Additionally, the fellow learned how to apply the principles and practices of NP EN ISO/IEC 17025:2018 and NP EN ISO 15189:2014 to molecular and agglutination methods.

## **5. Public health microbiology management**

### ***Project One Health European Joint Programme simulation exercise (OHEJP SimEx)***

Supervisor: Mónica Oleastro

Intersectoral collaboration is an essential component of the One Health approach, which recognises the interconnectedness of the health of human, animals, and the environment. The OH European Joint Programme (OHEJP) developed a national foodborne outbreak table-top simulation exercise (SimEx) to practice the OH capacity and interoperability across public health, animal health and food safety sectors, improving OH preparedness to future disease outbreaks. For the implementation of the OHEJP SimEx, each country designated a National (NEL) and Local Exercise Leader (LELs) to prepare its participation in the exercise. NEL/LELs identified participants from the three sectors (Public Health, Animal Health, and Food Safety) with different competencies that are normally involved in foodborne outbreaks in Portugal: epidemiologists (Public Health, Animal Health, Food Safety), laboratory personal (Public Health, Animal Health), food-borne disease experts, veterinaries, and public health doctors working at national, central, and local levels. The OHEJP SimEx took place in June 2022, at the National Institute of Agrarian and Veterinary Research (INIAV), Oeiras. From a One Health perspective, the SimEx exercise allowed the identification of several strengths and weakness regarding the roles and functions of available systems, the constraints of existing legislation, the importance of harmonisation and data sharing, and the creation of common main messages adapted to each target sector. There is still a long way to go to ensure cooperation among Public Health, Animal Health, and Food Safety sectors, as a One Health approach relies not only on the awareness of 'field experts' but also political and organisational willingness and commitment.

Role: The fellow took part in the SimEx Project as LEL at INSA (Public Health sector). Sandra Cavaco Gonçalves, a researcher at INIAV, held the positions of both NEL (all sectors) and LEL at INIAV (Animal Health sector). The fellow's main responsibilities included planning and facilitating INSA's involvement, supporting the NEL, acting as a liaison between the SimEx Team/INSA and NEL/INSA, collaborating with the local evaluator, identifying individuals and filling positions in the outbreak team, and preparing them for the exercise. The fellow worked as a facilitator, moderator, and supervisor during the conduct of the exercise, co-wrote the final report (Publications, other reports section 7.1.2, no. 3), and promoted scientific dissemination activities (e.g., workshops) and presented the results at national conferences (Conference presentations, section 7.2, nos. 8 and 9). In addition, the fellow was responsible for manuscript drafting and submission in peer-reviewed journal as first and corresponding author (Publications, manuscript section 7.1.1, no. 8).

### ***Observer at CESP Weekly Round Table meetings***

RONDA (Reunião sobre Observações, Notícias, Dados e Alertas) is an online meeting organised by the Public Health Emergencies Operations Centre (CESP), Portuguese Directorate General of Health (Direção-Geral da Saúde, DGS), as part of its coordination and early warning detection competencies. These meetings serve as a platform for different stakeholders, experts, and authorities to exchange updates, analyse current situations at both national and international level, and coordinate responses to public health emergencies in Portugal. From October 2022 to June 2023, the fellow attended the RONDA weekly meetings as an observer. The main goal was to enhance the comprehension of the meetings' importance within the field of public health and their role in effectively handling infectious threats at a national level. These meetings serve as a proactive approach to risk management and allow organisations to take timely actions to mitigate the negative impacts of risks and maintain resilience in the face of challenges.

Role: The fellow attended RONDA meetings as an observer and summarized her experience in a report.

### ***Educational outcome***

During the fellowship, all projects and activities included aspects of public health microbiology management competence. The fellow gained experience in establishing networks and contacts, communication within and between multidisciplinary work teams, research collaboration, time and team management, and effective communication with diverse target audiences.

## 6. Teaching and pedagogy

### *Course Field Epidemiology - outbreak investigation*

The EpiSurtos courses occurred at Lisbon's National Institute of Health Doutor Ricardo Jorge (INSA) on June 29-30, 2022 (9<sup>th</sup> edition), June 14-15, 2022 (Special edition for Public Health Doctors, EpiSurtos PHD 2022), June 22-23, 2023 (10<sup>th</sup> edition), and 6-7 July 2023 (EpiSurtos PHD 2023). EpiSurtos 9<sup>th</sup> and 10<sup>th</sup> editions targeted a broad audience of health professionals from various sectors (human and animal health, food safety) in public and private institutions. EpiSurtos PHD aimed at Public Health Residents from diverse Regional Health Administrations. The training included theory (fundamental epidemiology and outbreak concepts) and practice (hands-on participation in simulating the investigation of three outbreaks). In all EpiSurtos editions, a lecture titled 'The 10 Steps of an Outbreak Investigation' was delivered by the fellow. During this lecture, participants were actively encouraged to engage through real-time questions and answers, as well as by sharing their own experiences with outbreak investigations. The fellow also took on the role of a facilitator for three different case studies. In one of these case studies, focused on the 'Cholera Outbreak in India', the fellow updated the clarity of microbiological concepts. This included a brief presentation on 'The Laboratory Role in Outbreak Investigations'. At the conclusion of the training activity, an anonymous questionnaire was distributed to participants via email.

### *Public Health Specialisation Course (CESP)*

CESP is part of the post-graduate medical training programme in Public Health. The teaching activity was given as part of the 'Outbreak and Cluster Investigation and Control' module. The training comprised two parts: a theoretical component, with the presentation of the role of the laboratory in an outbreak; and a second, a practical component with the simulation of the investigation of an outbreak with the active intervention of the participants. The fellow delivered a lecture on 'Identification and characterization of microorganisms;' and acted as a facilitator for the case study 'An outbreak of gastroenteritis in Kalundborg, Denmark'. This case study was adapted and translated by the fellow for the teaching activity from the one presented at the EPIET/EUPHEM Introductory course 2021. This teaching activity took place at the Instituto de Saúde Pública da Universidade do Porto (ISPUP), in Oporto, on the 1 October 2022.

### *Supervisor of a Master student*

The fellow was the supervisor of a Master student, from the Faculty of Sciences of the University of Lisbon, with a thesis entitled: 'Characterization of antibiotic resistance in strains isolated from different environmental reservoirs'. The aim of the thesis that took place at the National Reference Laboratory for Antibiotic Resistance and Healthcare-Associated Infections (INSA) was to characterize the antibiotic resistance in strains isolated from different aquatic environmental reservoirs. Overall, the study demonstrated the diversity of antibiotic resistance genes and mobile-genetic elements within *Enterobacter cloacae* complex strains isolated from different aquatic environments, confirming that they can act as a reservoir of antibiotic resistant bacteria that can be pathogenic to humans. The study's findings led to the suggestion of establishing a monitoring and surveillance network in various environmental reservoirs beyond just water, in order to prevent the spread of antibiotic resistance.

### *OHEJP SimEx*

As LEL of the OHEJP SimEX Project (see section 5), the fellow was responsible for preparing the training audience for the exercise. Together with the NEL, she conducted a session to introduce the participants to the importance of simulation exercises, the tabletop format used, and the objectives and goals of the SimEx exercise. During this session, everyone introduced themselves, which provided an opportunity to understand each person's role before the exercise began. The fellow also gave a brief overview of Next-Generation Sequencing and the FoodChainLab tool because it would be helpful for the participants to be familiar with these approaches throughout the exercise. Throughout the SimEx, the fellow assumed the roles of a facilitator and exercise leader. Once the exercise concluded, a survey was emailed to all participants (including the training audience, NEL, LEL, and evaluators). The participants in the survey expressed interest in using the FoodChainLab tool in their future work. As a result, the fellow and the NEL collaborated with the German Federal Institute for Risk Assessment (BfR) and with the European Food Safety Authority (EFSA) to organise a face-to-face workshop. The purpose of the workshop was to provide training on how to use the FoodChainLab tool effectively (programme available at <https://foodrisklabs.bfr.bund.de/foodchain-lab-training-oeiras-portugal-february-2023/>). The workshop occurred on the 22-23 February 2023, at the Instituto Nacional de Investigação Agrária e Veterinária (INIAV) in Lisbon, Portugal.

### *Educational outcome*

The fellow taught different types of audience which required specific adaptations. The fellow developed communication and pedagogical skills, as she had to identify training needs, plan and organise seminars and workshops, prepare teaching material and case studies, and give lectures to persons from diverse backgrounds. Furthermore, the fellow's skills and knowledge in basic concepts of outbreak investigation and epidemiology, which were learned during the fellowship, were significantly enhanced through the teaching activities.

## 7. Communication

### 7.1 Publications related to the EUPHEM fellowship

#### 7.1.1 Manuscripts published in peer-reviewed journals

1. Perez Duque M, Ribeiro S, Martins JV, Casaca P, Leite PP, Tavares M, Mansinho K, Duque LM, Fernandes C, Cordeiro R, Borrego MJ, Pelerito A, de Carvalho IL, Nuncio S, **Manageiro V**, Minetti C, Machado J, Haussig JM, Croci R, Spiteri G, Casal AS, Mendes D, Souto T, Pocinho S, Fernandes T, Firme A, Vasconcelos P, Freitas G. Ongoing monkeypox virus outbreak, Portugal, 29 April to 23 May 2022. *Euro Surveill.* 2022; 27(22):2200424. <https://doi.org/10.2807/1560-7917.ES.2022.27.22.2200424>
2. Minetti C\*, **Manageiro V\***, Ourique M\*, Lucas M, Vicente A, Roque C, Coelho A, Oleastro M, de Sousa R, Rodrigues N. Epidemiological and genetic characterization of Norovirus outbreaks occurred in LVT region, Portugal 2017-2022. *In preparation.*
3. Sá R, Lopes C, Fonseca A, Almeida S, Roque J, Mendes PM, Gonçalves I, Ferreira MJ, Almendra T, das Neves N, Sousa J, Matos C, Fernandes A, Júnior A, Queiroz C, Coelho A, Maia C, Correia CB, **Manageiro V**, Minetti C, de Sousa R, Correia FH, Bodião J, Guerreiro, A.C. Challenges of investigating a large-scale outbreak of norovirus gastroenteritis in a hotel, Algarve, Portugal 2022. *Submitted to Archives of Virology.*
4. **Manageiro V**, Cano M, Iglesias C, Reis L, Vieira P, Teixeira A, Martins C, Machado J, Paiva JA, Caniça M. Outbreak of carbapenemase-producing *Enterobacterales* associated with environmental reservoirs, Portugal, 2022. *Submitted to Journal of Hospital Infection.*
5. **Manageiro V**, Matos R, Palminha P, Cortes-Martins H, Nunes B, de Sousa R. Tracking epidemiological shifts in hepatitis A in Portugal: a comparison of seroprevalence between two nationwide surveys 2001-2002 and 2015-2016. *Submitted to Eurosurveillance.*
6. **Manageiro V\***, Borges V\*, Rodrigues, R., Bettencourt, C., Silva, C., Gomes, J.P., Gonçalves, P. Microevolution and spatiotemporal dynamics of *Legionella pneumophila* ST1905 in Portugal, 2014-2022. *Submitted to Emerging Infectious Diseases.*
7. Borges V\*, **Manageiro V\***, (...), Gomes JP, de Sousa R. Genomic profiles of *Rickettsia conorii* strains isolated from patients with Mediterranean spotted fever, 1994-2022, Portugal. *In preparation.*
8. **Manageiro V**, Caria A, Furtado C, SimEx Portuguese Team, Botelho A, Oleastro M, Gonçalves SC. Intersectoral collaboration in a One Health approach: lessons learned from a country-level simulation exercise. *Submitted to One Health.*

\* Co-first authorship

#### 7.1.2 Other reports

1. **Outbreak Investigation report:** Norovirus outbreak in a long-term care facility, Portugal, 2022; completed in May 2023.
2. **Outbreak Investigation report:** Outbreak of carbapenemase-producing *Enterobacterales* in Braga, Portugal, 2022; completed in March 2023 [internal report, in Portuguese].
3. **Report** on SimEx Conduction in Portugal, 21<sup>st</sup> to 23<sup>rd</sup> of June 2022; completed in December 2022.

### 7.2 Conference presentations

1. Ourique M\*, Minetti C, **Manageiro V**, Vicente A, Roque C, Coelho A, Oleastro M, de Sousa R, Rodrigues N. Epidemiological investigation of a Norovirus GII.4 Sydney [P16] variant outbreak in a long-term care facility in Portugal, 2023. Congresso de Epidemiologia SEE/APE, 5-8 September 2023, Porto, Portugal. *Oral Communication.*
2. **Manageiro V\***, Borges V, Rodrigues R, Bettencourt C, Silva C, Gomes JP, Gonçalves P. 2023. Microevolution and spatiotemporal dynamics of *Legionella pneumophila* ST1905 in Portugal, 2014-2022. Congresso Nacional de Saúde Pública, 15-16 June 2023, Lisbon, Portugal. (Best Oral Communication Prize) *Oral Communication.*
3. **Manageiro V\***, Matos R, Palminha P, Cortes-Martins H, Nunes B, de Sousa R. 2023. Tracking epidemiological shifts in hepatitis A in Portugal: a comparison of seroprevalence between two nationwide surveys 2001-2002 and 2015-2016. 33<sup>rd</sup> European Congress of Clinical Microbiology & Infectious Diseases (ECCMID), 15-18 April 2023, Copenhagen, Denmark. P2824. *Paper poster presentation.*
4. **Manageiro V\***, Cavaco S\*. 2022. "Colaboração intersectorial numa abordagem One Health – Resultados de exercícios a nível nacional", Faculty of Veterinary Medicine of the University of Lisbon, 3 November 2023, Lisbon, Portugal. *Invited Oral Communication.*

5. **Manageiro V\***, Cavaco S\*. 2022. "Colaboração intersetorial numa abordagem One Health – Resultados de exercícios a nível nacional", National Congress of Public Health Medical Doctors (CNMSP), 3 November 2023, online. *Invited Oral Communication*.
6. Cavaco S\*, **Manageiro V**. 2022. "One Health EJP SimEx experiences across Europe: Portuguese Results", One Health EJP Joint SimEx/Dissemination Workshop: A One Health Simulation Exercise as a roadmap for future foodborne outbreak preparedness, 6 December 2022, online. *Invited Oral Communication*.
7. **Manageiro V\***, Ferreira E, EARS-Net-Portugal participants, Caniça M. 2022. Trends in antibiotic resistance in Portugal: analysis of surveillance data from 2015 to 2020. European Scientific Conference on Applied Infectious Disease Epidemiology (ESCAIDE), 23-25 November 2022, Stockholm, Sweden. *Oral Poster Presentation*.
8. **Manageiro V\***, Matos R, Palminha P, Cortes-Martins H, Nunes B, de Sousa R. Tracking epidemiological shifts in hepatitis A in Portugal: a comparison of seroprevalence between two nationwide surveys 2001-2002 and 2015-2016. Abstract submitted to European Scientific Conference on Applied Infectious Disease Epidemiology (ESCAIDE) 2022; not accepted.
9. Ourique M<sup>#</sup>, **Manageiro V<sup>#</sup>**, Minetti C<sup>#</sup>, Vicente A, Roque C, Coelho A, Oleastro M, Rodrigues N, de Sousa R, and the Outbreak Investigation Team. Epidemiological investigation of a Norovirus GII.4 Sydney variant outbreak in a long-term care facility in Portugal, 2022. Abstract submitted to European Scientific Conference on Applied Infectious Disease Epidemiology (ESCAIDE) 2022; not accepted. <sup>#</sup>Equal contribution
10. Sá R\*, Lopes C, Fonseca A, Almeida S, Roque J, Mendes PM, Gonçalves I, Ferreira MJ, Almendra T, das Neves N, Sousa J, Matos C, Fernandes A, Júnior A, Queiroz C, Coelho A, Maia C, Correia CB, **Manageiro V**, Minetti C, de Sousa R, Correia FH, Bodião J, Guerreiro, A.C. Challenges of investigating a large-scale outbreak of norovirus gastroenteritis in a hotel, Algarve, Portugal 2022. Abstract submitted to the 16th European Public Health Conference (EPHC) 2023; not accepted.

\* Presenting author(es)

### 7.3 Other presentations

1. **Manageiro V\*** 2021. Kick-off meeting for EUPHEM: presentation to introduce the fellow's background to potential project supervisors from the different laboratories of the Department of Infectious Diseases and Department of Epidemiology, 17 September 2021, INSA, Lisbon, Portugal.
2. **Manageiro V\*** 2021. OneHealth: Microbes are everywhere. Oral presentation during EUPHEM Introductory course Part 1, 20 September to 8 October 2021, online.
3. **Manageiro V\*** 2022. Trends in antibiotic resistance in Portugal: analysis of surveillance data from 2015 to 2020. Oral presentation at Project Review Module 2022, 29 August to 02 September 2022, Lisbon, Portugal.
4. **Manageiro V\*** 2022. Tracking epidemiological shifts in hepatitis A in Portugal: a comparison of seroprevalence in 2001-2002 and 2015-2016. Oral presentation at Project Review Activity – EUPHEM/EPIET Introductory Course 2021 part 2, 20-29 April 2022, Spetses, Greece.
5. MLCPH Group 1 (Hommes F, Laisnez V, **Manageiro V**, Otte im Kampe E, Roy A, Schmidt K, Stratil J\*, van Ewijk K). 2023. Competences, skills, and personal characteristics for a public health leader to be successful in a crisis period. Oral presentation at Management, Leadership and Communication in Public Health (MLCPH) Module 2023, 8-12 May 2023, Stockholm, Sweden.
6. **Manageiro V\*** 2023. Genomic profiles of *Rickettsia conorii* strains isolated from patients with Mediterranean spotted fever, 1994-2022, Portugal. Oral presentation at Project Review Module 2023, 28 August to 01 September 2023, Lisbon, Portugal.

\* Presenting author(es)

## 8. EPIET/EUPHEM modules attended

1. **Introductory Course Part 1**, 20 September to 8 October 2021, online
2. **Introductory Course Inject Day 1 on Phylogeny & Whole genome sequencing**, 20 October 2021, online
3. **Introductory Course Inject Day 2 on Operational Research**, 27-28 October 2021, online
4. **Introductory Course Inject Day 3 on Data Collection and Management**, 10-11 November 2021, online
5. **Outbreak Investigation**, 6-10 December 2021, online
6. **Biorisk and Quality Management**, 11-13 January 2022, online

7. **Multivariable analysis**, 14-18 March 2022, online
8. **Multivariable analysis – Cox Regression Inject Day**, 30 March 2022, online
9. **Introductory Course Part 2** (including Project Review Activity), 20-29 April 2022, Spetses, Greece
10. **Rapid Assessment and Survey Methods**, 6-10 June 2022, Stockholm, Sweden
11. **Project Review 2022**, 29 August to 2 September 2022, Lisbon, Portugal
12. **Time Series Analysis**, 7-11 November 2022, Utrecht, The Netherlands
13. **Qualitative Research Virtual Inject Days**, 31 January, and 3 February 2023, online
14. **Vaccinology**, 13-17 February 2022, online
15. **Management, Leadership and Communication in Public Health**, 8-12 May 2023, Stockholm, Sweden
16. **Project Review 2023**, 28 August 2023 to 1 September 2023, Lisbon, Portugal

## 9. Other training

1. Rotation in the National Reference Laboratories from the Department of Infectious Diseases (INSA), 13 October 2021 to 16 February 2022, Lisbon, Portugal
2. European Scientific Conference on Applied Infectious Disease Epidemiology (ESCAIDE) 2021, 16-19 November 2021, online
3. Webinar “O papel do Laboratório no diagnóstico de Tuberculose” (The Laboratory role in the diagnosis of Tuberculosis), 6 December 2021, online
4. ECDC e-learning courses “How to design a Functional Exercise (FX)” and “How to design a Table-top Exercise (TTX)”, 25 January 2022, ECDC Virtual Academy (EVA), online
5. Dissemination Workshop on Improving One Health Preparedness to (re)emerging Infectious Threats, OneHealth EJP, 25 March 2022, online
6. One Health EJP ASM Satellite Workshop: Diagnostics workshop; mobile detection platforms for One Health diagnostics applications, OneHealth EJP, 14 April 2022, online
7. Public Health Microbiology lecture series by EUPHEM Fellows (Virology and Immunology), 12 May and 19 May 2022, online
8. Mass gathering WHO course, 19 May 2022, online
9. United Nations “BSAFE security awareness training” course, 20 May 2022, online
10. Epidemic Intelligence e-learning course, 30 May 2022, online
11. Monkeypox: Introductory course for African outbreak contexts, OpenWHO, 22 May 2022, online
12. ESCMID/IDSA joint web symposium on Monkeypox, 24 May 2022, online
13. ESGEM/ESCMID Webinar “From the Present to the Future of WGS Surveillance for Food Safety”, 14 June 2022, online
14. EFWISG/ESCMID Webinar “Genomic surveillance of viral pathogens: are we prepared for emergent global threats across One Health”, 20 September 2022, online
15. EFWISG/ESCMID Webinar “Comparison of new diagnostic techniques multiplex PCR and WGS enteric infection surveillance with conventional diagnostic procedures”, 20 September 2022, online
16. Introduction to Git/Github, presented by Liza Coyer (EPIET C2021) 21 October 2022, online

17. EAN webinar "Xenomonitoring and surveillance: Using mosquitoes to find (and control) pathogens", presented by Corrado Minetti (EUPHEM C2021), 26 October 2022, online
18. ESGPHM/ESCMID Webinar "New roles of microbiology laboratories in public health surveillance and epidemic response", 27 October 2022, online
19. European Antibiotic Awareness Day 2022 event, 17 November 2022, online
20. EAN mini-module on Media and Infodemic Management, 21-22 November 2022, Swedish Public Health Agency, Stockholm, Sweden
21. European Scientific Conference on Applied Infectious Disease Epidemiology (ESCAIDE) 2022, 23-25 November 2022, Stockholm, Sweden
22. FoodChain-Lab Workshop, 22-23 February 2023, Instituto Nacional de Investigação Agrária e Veterinária (INIAV), Oeiras, Portugal
23. EPI-WIN webinar: Cholera outbreaks: current situation and what we are doing to address it, WHO, 15 March 2023, online
24. Pre-ECCMID (European Congress of Clinical Microbiology & Infectious Diseases) Day on AMR/AMS: Emerging resistance mechanisms spreading across sectors, 16 March 2023, online
25. 14<sup>th</sup> Workshop of the Vector Surveillance Network - REVIVE, 13 April 2023, online
26. 33<sup>rd</sup> European Congress of Clinical Microbiology & Infectious Diseases, 15-18 April 2023, Copenhagen, Denmark
27. EAN Mobile Laboratory Webinar, 04 May 2023, online
28. Congresso Saúde Pública, 15-16 June 2023, Lisbon, Portugal
29. Field Epidemiology Course - Mass Events, 17-18 July 2023, INSA, Lisbon, Portugal
30. European Scientific Conference on Applied Infectious Disease Epidemiology (ESCAIDE) 2023, 22-24 November 2023, Barcelona, Spain

## 10. Other activities

### EUPHEM representative for Cohort 2021

At the beginning of the fellowship, the fellow was nominated cohort 2021 EUPHEM Ms-Track representative. In this role, she was engaged in various tasks, including obtaining views and ideas of fellows; gathering and collating fellows' issues or concerns to contribute to the ongoing enhancement of the EPIET and EUPHEM fellowship; supporting routine fellowship activities (e.g. fellow surveys, organization of meetings and social activities). Additionally, the fellow took part in multiple meetings alongside the other representatives from the 2020 to 2022 cohorts, namely the Active Supervisors and Training Site Forum, National Focal Points for Training and Training Site Forum, and the EPIET Alumni Network (EAN).

### EPIET Alumni Network (EAN) activities

The fellow, as a member of the EAN community, took on a volunteer role in the organisation of the one-day celebration of over two decades of EAN and more than twenty-five years of EPIET (EAN 25+). This event took place in the Swedish Public Health Agency in Stockholm on Saturday 26 November 2022, one day after ESCAIDE 2022. It was a get together event to reflect around the past, present, and future of EPIET and field epidemiology in Europe. It gathered about 150 alumni from all the previous and current cohorts. Afterwards, the fellow contributed to the 'EPIET Alumni Network – Winter 2023 Newsletter', by collaborating on its edition and co-authoring a news article covering the EAN 25+ event (available at <https://epietalumni.net/eat-newsletters/>).

### Newspaper

A journalist from the Portuguese daily national newspaper 'Público', based in Lisbon, Portugal, observed the ninth edition of EpiSurtos (refer to section 6). This observation led to a news article titled 'Caça-surtos: aprender a ser detective na Saúde' ('Outbreak hunter: learning to be a detective in health'), which was published on 17 July 2022 in both digital and paper formats (available at <https://www.publico.pt/2013079>). The news article recounted the narrative of the EpiSurtos course and included the fellow's role as a course facilitator.

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