Summary of work activities
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European Public Health Microbiology Training Programme (EUPHEM), 2019 cohort

Background

According to the European Centre for Disease Prevention and Control (ECDC) Advisory Group on Public Health Microbiology ('national microbiology focal points'), public health microbiology is a cross-cutting area that spans the fields of human, animal, food, water and environmental microbiology, with a focus on human population health and disease. Its primary function is to improve health in collaboration with other public health disciplines, in particular epidemiology. Public health microbiology laboratories play a central role in detection, monitoring, outbreak response and the provision of scientific evidence to prevent and control infectious diseases.

European preparedness in responding to new infectious disease threats requires a sustainable infrastructure capable of detecting, diagnosing and controlling infectious disease problems, including the design of control strategies for the prevention and treatment of infections. A broad range of expertise, particularly in the fields of epidemiology and public health microbiology, is necessary to fulfil these requirements. Public health microbiology provides experts in all relevant communicable diseases at the regional, national and international level with the tools they need to mount rapid responses to emerging health threats. This enables them to plan appropriate prevention strategies, assess existing prevention disciplines, develop microbiological guidelines, evaluate/produce new diagnostic tools, assess risks from microbes or their products and provide pertinent information to policy makers from a microbiological perspective.

According to Articles 5 and 9 of ECDC's founding regulation (EC No 851/2004) 'the Centre shall, encourage cooperation between expert and reference laboratories, foster the development of sufficient capacity within the community for the diagnosis, detection, identification and characterisation of infectious agents which may threaten public health' and 'as appropriate, support and coordinate training programmes in order to assist Member States and the Commission to have sufficient numbers of trained specialists, in particular in epidemiological surveillance and field investigations, and to have a capability to define health measures to control disease outbreaks'.

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' which is why 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.

This report summarises the work activities undertaken by Justine Schaeffer, cohort 2019 of the European Public Health Microbiology Training Programme (EUPHEM) at the Agentur für Gesundheit und Ernährungssicherheit (Austrian Agency for Health and Food Safety, AGES), Vienna, Austria.

The fellow graduated with a Masters degree in fundamental Virology from the École Normale Supérieure, the Université Paris Diderot and the Institut Pasteur (Paris, France). She then did a PhD on the immune response to Lassa virus (Institut Pasteur, Lyon, France), working in a BSL-4 facility. During her PhD, she had the opportunity to take part in two field missions, carrying out Ebola diagnosis in Guinea during the West African Ebola epidemic (2014-2015). This experience led her to consider leaving academia for public health. After her PhD, she went back into the field as a lab manager for the NGO the Alliance for International Medical Action (ALIMA). She was responsible for the rehabilitation, management, and improvement of a Lassa fever diagnosis laboratory in a Nigerian hospital. She returned to Europe to start the EUPHEM training at the Austrian Agency for Health and Food Safety (AGES).

All EUPHEM activities aim to address different aspects of public health microbiology and underline the various roles of public health laboratory scientists within public health systems.

Methods

This report accompanies a portfolio demonstrating the competencies acquired during the EUPHEM fellowship by working on various projects, activities and theoretical training modules.

Projects included epidemiological investigations (outbreaks and surveillance); applied public health research; applied public health microbiology and laboratory investigation; bio-risk management; quality management; teaching and public health microbiology management and the summary and communication of scientific evidence and activities with a specific microbiological focus.

The outcomes include publications, presentations, posters, reports and teaching materials prepared by the fellow. The portfolio presents a summary of all work activities conducted by the fellow, with the exception of those prohibited for reasons of confidentiality.

Results

The objectives of these core competency domains were achieved partly through project or activity work and partly through participation in the training modules. Results are presented in accordance with the EUPHEM core competencies, as set out in the EUPHEM scientific guide[1].

1. Epidemiological investigations

1.1. Outbreak investigations

1.1.1. SARS-CoV-2 diagnosis at AGES and integration in Austrian diagnosis capacities
Supervisors: Daniela Schmid, Alexander Indra, Peter Hufnagl

Following its emergence in China, SARS-CoV-2, the causing agent of COVID-19, has spread worldwide. In the first half of 2020, Europe was particularly affected, but with great differences between countries. In Austria, a solid preparedness plan was developed as early as January 2020. It partially explained the Austrian success in controlling its COVID-19 epidemic.

In Vienna, many actors have been involved in this preparedness, and in the response later on. Since January 2020, AGES' Molecular Microbiology Department was one of the four laboratories in Vienna implementing SARS-CoV-2 diagnostic by RT-qPCR. In addition to their participation in the diagnostic efforts, AGES also supported the implementation of SARS-CoV-2 testing in other laboratories throughout Austria. In 2021, concern was growing in Europe regarding SARS-CoV-2 variants (lineage B.1.1.7, B.1.351 and P.1). If monitoring of such variants was first organised using specific PCR, sequencing-based methods provided more precise surveillance of SARS-CoV-2 genetic diversity. AGES Molecular Microbiology was again in the first line of national efforts, implementing amplicon-based whole genome sequencing of SARS-CoV-2.

Role of the fellow: as early as January 2020, the fellow was responsible for the implementation of SARS-CoV-2 diagnostic at AGES. She also collaborated with other Viennese laboratories to coordinate diagnostic efforts. This included timelines of decision-making, implementation of diagnostic techniques, adaptations of the workflow during the different stages of epidemic and support to laboratories throughout the country willing to implement SARS-CoV-2 diagnostic. To keep herself updated on the fast-evolving knowledge on COVID-19, the fellow had to perform a regular literature watch and attended several webinar on topics such as clinical presentation and management,

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[1] European Centre for Disease Prevention and Control. European public health training programme. Stockholm: ECDC; 2013. Available from: http://ecdc.europa.eu/en/publications/Publications/microbiology-public-health-training-programme.pdf

laboratory preparedness and vaccine development. In 2021, she organised, tested and validated SARS-CoV-2 sequencing at AGES.

1.1.2. Cross-border outbreak of Listeria monocytogenes Ny9
Supervisors: Ariane Pietzka, Franz Allerberger

In November 2020, two case of invasive listeriosis were reported to the National Reference Centre (NRC) in Austria. Microbiological characterisation of the *Listeria monocytogenes* clinical isolates showed that both strains were genetically similar (0 allelic differences in cgMLST), pointing at a common-source outbreak. On 2 December 2020, an outbreak investigation was started by the Austrian Agency of Health and Food Safety to identify the vehicle of infection and prevent further contaminations.

On 16 November 2020, an Epidemic Intelligence System (EPIS) notification reported an outbreak of *L. monocytogenes* Ny9 in Germany. Comparison between the two Austrian isolates and the German outbreak strain showed 0 allelic differences in cgMLST, suggesting that they belonged to the same outbreak. On 4 December 2020, this cross-border *L. monocytogenes* Ny9 outbreak included 30 cases in Germany, two cases in Austria, one case in Denmark, and one case in Switzerland. Collaboration with the institutes in charge of the outbreak investigation in Germany, Denmark, and Switzerland was initiated to share information and conclusions regarding the outbreak investigation.

In Austria, the two cases were interviewed about their food consumption by the local public health authorities. Both reported consumption of smoked trout fillet from the same brand and purchased in the same supermarket chain. This food item was mentioned in a Rapid Alert System for Food and Feed (RASFF) notification, as *L. monocytogenes* had been detected in this product. Whole genome sequencing analysis of the RASFF isolate showed high similarity with the outbreak strain (1 allelic difference). This finding supported the hypothesis that smoked trout was the vehicle of contamination for the two Austrian cases.

At the time of the investigation, a silent recall of smoked trout had already been conducted by the supermarket chain headquarter. On 3 December 2020, AGES issued a warning on this product, informing the public that incriminated products still present at home should not be consumed under any circumstances. The next date, the supermarket chain issued an official recall of smoked trout fillets from the incriminated brand. No further cases have been reported since. The Danish manufacturer of the incriminated product has been informed and investigated by the Danish health authorities. On the Austrian side, the investigation was closed at the end of December 2020, even though the NRC for *L. monocytogenes* remained vigilant regarding the report of additional cases.

Role of the fellow: she was involved in this outbreak investigation under the supervision of the head of the NRC. She helped with some of the environmental investigations, gathered information on the timeline of events related to this outbreak for the outbreak investigation report, and exchanged with RKI regarding the results of their investigation.

1.1.3. Training modules related to the assignment/projects
EPIET/EUPHEM Introductory Course: the basics of outbreak investigation were covered during this course. Learning objectives included the principles and the 10 steps of outbreak investigation, questionnaire design, data collection and entry, analysis of the study results and formulation of recommendation. Theoretical concepts were taught through lectures and applied during practical group works, including several case studies

Outbreak investigation module: more in-depth insight into outbreak investigation was obtained during this module. Many examples of successful and not so successful outbreak investigations were given. Questionnaires were designed and tested by the fellows. Two outbreaks were analysed in case studies, using R to perform descriptive and univariate analysis of the data. For one of these case studies, an outbreak investigation report was written in groups. A workshop introduced the fellows to epidemic intelligence.

Multivariate analysis module: during this module, fellows learned how to perform multivariate analysis, which can be applied for both outbreak investigation and public health research. Theoretical and practical sessions taught fellows the different type of models for multivariate analysis, how to choose which model to use, how to construct the model, evaluate the most relevant one and interpret the model's parameters.

Vaccinology module: this module highlighted the specificities of vaccine preventable diseases outbreaks, in term of investigation, communication and control measures. The fellows also learned how to assess advert effects following vaccination using investigation techniques similar to outbreaks.

Educational outcome

The fellow discovered the fundamental principles of outbreak investigation during the fellowship modules. She then was able to apply them through the preparedness, detection, and investigation of outbreaks at her training sites. In addition to the microbiological and epidemiological aspects, collaboration, communication and management

were important parts of the of the outbreak investigations. Both the COVID-19 response and the Listeria outbreak in which the fellow was involved included national and international levels.

1.2. Surveillance

1.2.1. Setting up a surveillance system for Hantaviruses in rodents
Supervisors: Georg Duscher, Franz Allerberger

Hantaviruses are rodent-borne viruses able to infect humans. Clinical features vary from asymptomatic to very severe. Overall, even though the clinical burden of Hantaviruses in Europe is still low, their emerging potential is rising. In Austria, global warming is anticipated to have a huge impact on forest (with a switch from pine trees to oaks mainly), changes that would increase rodent populations. In the case of Hantaviruses, such changes in rodent ecology would have a major influence, with a predicted increase in human cases. Most Austrian Hantavirus infections are currently reported in Styria. In this region, assessing Hantavirus circulation in rodents is of major importance to better understand the routes of contamination and to recommend adapted control measures. Therefore, AGES was interested in setting up a surveillance system for Hantaviruses in rodent, starting in Styria.

This rodent surveillance system was coordinated by AGES in collaboration with the University Hospital for Internal Medicine of Graz and of the Veterinary Medicine University of Vienna. Briefly, participants were to be recruited at the University Hospital for Internal Medicine of Graz and given rodent sampling kits, including labelled traps. Successful traps would be transferred by AGES Graz to the Pathology department of the Veterinary Medicine University of Vienna, which would harvest organs from the rodents. Harvested organs would be tested at AGES by the fellow, who would then share results with the participant and the other stakeholders. If positive samples were found, they would be sent to the Virology Department of Vienna Medical University for sequencing.

Role of the fellow: the fellow participated in the design of this surveillance system and the recruitment of stakeholders. She also developed the documents needed for the surveillance system (description of the system, recruitment and consent form for the participants, follow-up of the collected samples, result communication). On behalf of AGES, she was responsible for the molecular detection of Hantaviruses in rodent samples, including the implementation of an adapted technique. She chose an RNA extraction with organ fragmentation, followed by a pan-Hantavirus nested PCR. She tested the workflow and wrote the standard operating procedure.

1.2.2. New typing method for Mycobacterium avium subspecies paratuberculosis surveillance
Supervisors: Werner Ruppitsch, Eva Sodoma, Michaela Altmann

Paratuberculosis, or Johne's disease, is a chronic and incurable infectious disease of domestic and wild ruminants, caused by *Mycobacterium avium subspecies paratuberculosis* (MAP). Mild cases have an important economic impact due to weight loss and declining milk production, and more severe cases can lead to fertility disorders and death. Paratuberculosis is listed by the World Organization for Animal Health (OIE) and must be reported. Although MAP zoonotic transmission is still being evaluated, it is pointed at as one of the co-factors for Crohn's disease. Therefore, surveillance of MAP fits into the One Health approach. It also matches the missions of AGES, which includes supporting the management of the Federal Ministries in matters of public health and animal health.

The National Reference Centre (NRC) for paratuberculosis at AGES Institute for Veterinary Medicine in Linz is responsible for MAP diagnosis and surveillance. As MAP is world-spread and cross-border, classification of the strains is important to identify the patterns of infection in livestock and track the chains of transmission. Therefore, it is essential to be able to conciliate different classification systems that might be used in other countries. By adding new typing method to the NRC panel, we strengthened its ability to compare Austrian MAP strains at an international level.

The NRC for paratuberculosis requested help from AGES molecular microbiology department to implement a new classification system. A SNP-based typing method published in 2016 by Leão & al. was adapted to the needs of the NRC. As MAP isolates are routinely sequenced, we used whole genome sequencing (WGS) data as input. We developed a bioinformatics workflow on the user-friendly tool Seqsphere+. This workflow extracted 14 target SNP from WGS data, and sequential analysis of these 14 SNP allowed to type the isolates. A final test on almost 300 sequences showed good performance of the workflow, with typing up to the clade level for almost 90% of the isolates. Further analysis of the sequences correlated workflow failure with lower sequence quality.

Therefore, this workflow could provide the NRC with a potent tool to type MAP isolates, especially if good WGS quality is achieved. In addition, the collaboration between the NRC and the molecular microbiology department resulted in valuable capacity building for both parties involved.

Role of the fellow: the fellow was the principal investigator of this collaboration on the side of AGES molecular microbiology department. She assessed the need of the NRC for paratuberculosis, developed the typing workflow and tested its efficiency. She also wrote detailed instructions on how to use these new tools and a final report summarising all the tests she ran.

1.2.3. High-level macrolides resistances in Campylobacter

Supervisor: Sandra Köberl-Jelovcan

Campylobacter, especially *C. jejuni* and *C. coli*, are one of the leading causes of foodborne diarrheal disease worldwide. Gastrointestinal symptoms are usually self-limiting, but antibiotic treatment is recommended for severe or prolonged illness and immunocompromised hosts. Macrolides are the drug of choice for human campylobacteriosis. Macrolide resistance in *Campylobacter* usually relies on point mutations in the 23S rRNA gene, but *Campylobacter* strains carrying *erm(B)* have been described. In this study, we investigated the genetic background of Austrian *Campylobacter* isolates resistant to erythromycin. *Campylobacter* strains isolated in Austria in 2016-2021 were characterised by whole genome sequencing (WGS). Antimicrobial resistance genes or mutations were identified using three different online tools: RGI from the Comprehensive Antibiotic Resistance Database, ResFinder from the Center for Genomic Epidemiology and NCBI AMR finder. Isolates were typed using core genome MLST.

48 *C. coli* (75%) and 16 *C. jejuni* (25%) strains, from humans (54) or poultry (10), were included in this study. Phylogenomic classification showed a strict segregation between *C. jejuni* and *C. coli* isolates, but not according to any other variable. Two potential genetic determinants of macrolide resistance were found, 23S rRNA point mutation A2075G or A2074T/C/G and Erm(B) rRNA methylase. For most of the *Campylobacter* isolates, high level of erythromycin resistance could be explained by 23S rRNA point mutations. However, three isolates carrying *erm(B)* were identified for the first time in Austria. All *erm(B)* genes were located in a multi-drug resistance genomic island including also aminoglycoside and tetracycline resistance genes. The detection of Campylobacter isolates carrying *erm(B)* is a source of concern given its association with other antimicrobial resistances. Also, the prevalence of such strains increased rapidly in Asian countries where it has been previously described. Monitoring the genetic determinants of macrolide resistances would allow to track the potential spread of *erm(B)*+ *Campylobacter* in Austria.

Role of the fellow: the fellow performed the genomic analysis, to classify the isolates by core genome MLST and identify know resistance genes. She compared the performances of the three analysis tools. She performed univariate analysis to measure the association between genomic and phenotypic AMR. She communicated the results with one manuscript in peer-reviewed journals and a poster presentation at an international conference.

1.2.4. Training modules related to the assignment/projects

EPIET/EUPHEM Introductory Course: the rationale behind surveillance, but also the structure and attributes of a surveillance system, were studied during this module. The different types of surveillance and their key components were the topic of several lectures. Through cases studies, the fellows also learned how to analyse and interpret surveillance data, and how to evaluate a surveillance system.

Outbreak investigation module: this module helped the fellow develop analytical skills that were designed for outbreak investigations but can also be useful for surveillance. In cases studies, the fellows learned to perform descriptive epidemiology using R. Data mapping was also covered, through a lecture and a practical exercise.

Time series analysis module: this module covered all the step to analyse surveillance data over time. Through lectures and practical exercises, fellows analysed the global trend and periodicity of time series, residual patterns and unusual events. They also used various periodical models to predicted further evolutions, compare two time series and evaluate interventions.

Rapid Assessment and Survey Methods module: this module covered the specificities of surveillance and surveys in challenging settings (low-resources, emergency response, refugee camps…). Participants went through simulation exercises on the detection, early response and sustained follow-up of an international public health emergency.

Vaccinology module: during this course, fellows learned about the specificities of surveillance systems for vaccine-preventable-diseases, through lectures and case studies.

Educational outcome

The combination of modules and projects gave to the fellow a global knowledge of every aspects of surveillance: evaluate the need for surveillance, set up a surveillance system, understand how surveillance works, evaluate a surveillance system and use surveillance data to inform public health measures. She collected data from both epidemiological and genomic surveillance to understand the specific patterns of diseases. She worked on a pilot project to set up a Hantavirus surveillance system in rodents, recruiting stakeholders and organising a functioning workflow.

2. Applied public health research

2.1. Sequencing of Austrian *Corynebacterium* isolates from 2010 to 2019

Supervisor: Werner Ruppitsch

Diphtheria is a vaccine-preventable disease with a high potential for re-emergence. Its main causative agents are *Corynebacterium diphtheriae* and *Corynebacterium ulcerans*, with some strains producing diphtheria toxin. From 2010 to 2019, 57 clinical *C. diphtheriae* strains were isolated in Austria, either from the respiratory tract or from skin infections. Six *C. ulcerans* strains were isolated in the same time frame. The aim of this study was to investigate the genetic diversity of these *C. diphtheriae* and *C. ulcerans* isolates using whole-genome sequencing.

Among the six *C. ulcerans* strains, four originated from skin infections in human and two from wild boars. The two animal isolates and 2/4 human isolates carried the diphtheria toxin gene. Genomic data were published on the NCBI database to increase the pool of *C. ulcerans* sequences, as at the time only 17 complete and 13 draft genomes were publicly available. Increasing the number of available sequences would support molecular epidemiology studies, therapeutic and vaccine development, and also research projects investigating the mechanisms of *C. ulcerans* carriage, transmission, and pathogenesis.

C. diphtheriae isolates were characterised by genome-wide comparisons using single nucleotide polymorphism analysis or core genome multilocus sequence typing. Genome data were also searched for antimicrobial resistance genes and genes involved in diphtheria toxin production. The genetic diversity among the isolates was high, with no clear distribution over time or place. *C. belfantii* isolates were separated from other strains and were strongly associated with respiratory infections (odds ratio = 57). Two clusters, limited in time and space, were identified. Almost 40% of strains carried resistance genes against tetracycline or sulfonamides, mostly from skin infections. Microbiological tests showed that 55% of isolates were resistant to penicillin but did not carry genes conferring β-lactam resistance. A diphtheria toxin gene with no non-synonymous mutation was found in three isolates only. This study showed that sequencing can provide valuable information complementing routine microbiological and epidemiological investigations. It allowed to identify unknown clusters, evaluate antimicrobial resistance more broadly, and support toxigenicity results obtained by PCR. For these reasons, *C. diphtheriae* surveillance could strongly benefit from the routine implementation of whole-genome sequencing.

Role of the fellow: as the principal investigator of this study, the fellow was in charge of data collection, analysis and publication. She performed the sequencing of the isolates, developed a core-genome MLST scheme for *C. diphtheriae* and *C. ulcerans*, and analysed WGS data. She also performed additional bioinformatics analysis, such as single nucleotide polymorphism analysis, alignment of diphtheria toxin/diphtheria toxin regulator genes and search of antimicrobial resistance genes. She collected clinical data and compared them with microbiological data. She wrote two manuscripts and handled their publication in peer-reviewed journals.

2.2. *Brucella melitensis* diversity in Austrian clinical and food samples

Supervisor: Werner Ruppitsch

Brucellosis is a zoonotic disease caused by *Brucella spp* and a major concern for livestock. Most human cases are caused by *B. melitensis* and clinical presentation is usually a mild febrile illness. However, treatment failure is frequent and more severe complications can occur. In Austria, every human brucellosis is investigated to determine whether it was imported from endemic areas or is the sign of an undetected autochthonous transmission.

For this study, 21 *B. melitensis* strains isolated in Austria between 2005 and 2019 were collected: 17 strains from 15 different patients and four strains from cattle. Whole genome sequencing combined with core-genome MLST analysis was used to characterise these strains. A cluster of seven isolates from 2018 (three human and four cattle isolates) was identified, with fewer than two allelic differences. They corresponded to the only Austrian *B. melitensis* outbreak that happened over the past 15 years.

The other 12 Austrian brucellosis cases were single cases, and geographical origins were available for 8/12. Genomic data was used to locate probable geographical origins and compared with the results of the epidemiological investigations. Austrian strains were compared with 67 published *B. melitensis* sequences available on NCBI. The result of genomic analysis matched for 7/8 cases with documented conclusion of the epidemiological investigation. Genome analysis also pointed to a possible geographical origin for three of the four cases with missing epidemiological data. Strains from six cases grouped together (<40 allelic differences) with 4/6 cases imported from the Balkans. Additional *B. melitensis* isolates from Serbian animals were analysed and grouped with this branch, suggesting frequent importation from Balkan countries to Austria.

Overall, this study highlighted the specificities of human brucellosis in Austria. It also underlined the value of whole genome sequencing as a tool to investigate brucellosis cases, allowing to identify and investigate outbreaks but also to support epidemiological investigation of imported cases. However, the reliability of such methods depends

on the number of strains for comparison, which can be challenging in low incidence countries. Increasing the availability of published sequences with documented geographical origins would help establishing genomic-based methods for investigating brucellosis cases.

Role of the fellow: the fellow was as principal investigator of this study. She analysed whole genome sequencing data, using single nucleotide polymorphism analysis, core genome MLST and antimicrobial resistance genes identification tools. She collected clinical and epidemiological data and compared them with microbiological data. She communicated the study results as a manuscript in a peer-reviewed journal and an oral presentation at an international conference.

2.3. Antimicrobial resistance genes in *Neisseria gonorrhoeae*

Supervisor: Werner Ruppitsch

Gonorrhoea is the second most common bacterial sexually transmissible infection in the EU and worldwide. The majority of cases are asymptomatic or very mild, but if untreated more severe symptoms can occur. Gonorrhoea is treated with antibiotics, however, the plasticity of *Neisseria gonorrhoeae*'s genome favours the acquisition and dispersion of antimicrobial resistance (AMR) genes. Since the 1940s, *N. gonorrhoeae* has developed or acquired AMR to all antimicrobials previously recommended as gonorrhoea empirical treatment. The Austrian reference centre for *N. gonorrhoeae* is located at AGES and is in charge of surveillance. *N. gonorrhoeae* strains diagnosed at the NRC or sent by partner institutions are sequenced. Combining genomic, epidemiological and clinical data, we investigated the population structure of Austrian *N. gonorrhoeae* and the genomic determinants of AMR.

Between 2016 and 2020, AMR rate was increasing for all tested antibiotics except penicillin. Combining AMR profiles with core genome MLST typing, we identified phylogenomic branches of interest. One branch had a high rate of azithromycin resistance, and all isolates carried a mosaic *mtr* gene. A distinct branch correlated with cephalosporin resistance, including all cefixime-resistant isolates and 24/32 isolates with reduced susceptibility to ceftriaxone. Overall, these results gave an overview of the Austrian *N. gonorrhoeae* population and highlighted the benefice of genomic surveillance for AMR monitoring.

As alternative therapies should be considered, we also looked at the potential efficacy of another antimicrobial, rifampicin. Indeed, rifampicin is one of the most potent and broad-spectrum antibiotics. Clinical trials showed the efficiency of rifampicin to treat gonorrhoea, but the potential of this treatment is conditioned by the pre-existence of rifampicin resistance in the *N. gonorrhoeae* population. We investigated rifampicin resistances in Austrian *N. gonorrhoeae* by searching for mutations in the *rpoB* genes which are known to confer rifampicin AMR. We found a rifampicin resistance rate of 30%, which is too high to recommend rifampicin as gonorrhoea treatment. As rifampicin resistance was correlated with other AMR, the use of rifampicin in combination therapy would also not be recommended.

Role of the fellow: the fellow gathered epidemiological and genomic data. She performed the genomic analysis, to classify the isolates by core genome MLST and identify know AMR genes. She performed time-series, univariate and multivariate analysis combining phenotypic AMR and genomic data. She communicated the results with two manuscript in peer-reviewed journals and two posters at international conferences.

2.4. Training modules related to the assignment/projects

EPIET/EUPHEM Introductory Course: during this module, fellows were acquainted with operational research, but also given statistical tools to perform descriptive and analytic epidemiology. The statistics covered epidemiology measures, confidence interval and statistical tests. Sample size calculation were also taught and applied through a case study. The different types of study design and their analysis were also the topic of several lectures and case studies, including measures of association, confounding and effect modification, bias and control group selection. During a research group project, the fellows had to design their own study, write the draft of a study protocol and present the project.

Outbreak investigation module: this module allowed fellows to get familiar with informatics tools to perform descriptive and analytical epidemiology. Data interpretation depending on the study design was also covered.

Management, Leadership and Communication in Public Health module: project management objectives, challenges and tips were provided during this module, through theoretical lectures and practical exercises. It included project planning, budgeting, time management, teamwork and efficient output communication.

Multivariate analysis module: during this module, fellows learned how to perform multivariate analysis, which can be applied to both outbreak investigation and public health research. Theoretical and practical sessions taught fellows the different type of models for multivariate analysis, how to choose which model to use, how to construct the model, evaluate the most relevant one and interpret the model's parameters.

Time series analysis module: this module covered all the step to analyse surveillance data over time. Through lectures and practical exercises, fellows analysed the global trend and periodicity of time series, residual patterns

and unusual events. They also used various periodical models to predicted further evolutions, compare two time series and evaluate interventions.

Rapid Assessment and Survey Methods module: the specific challenges of setting up a research project in a low-resources and unstable environment were discussed during this module.

Educational outcome

Thanks to several modules, the fellow was able to adapt what she learned in academic research to the specificities of public health. She was the principal investigator of several research projects, from the definition of the objectives to the communication of the results. She gained new bioinformatic and statistical skills to better analyse epidemiological and microbiological data. She presented the results of these research projects at scientific conferences and in peer-reviewed journals.

3. Applied public health microbiology and laboratory investigations

3.1. Standard protocol for enteroviruses whole genome sequencing

Supervisor: Birgit Prochazka

The Enterovirus genus consists in 4 species of enteroviruses (EV) and 3 species of rhinoviruses (HRV) infecting humans. Among EV, poliovirus (PV) is responsible for very severe acute flaccid paralysis in human. Non-polio EV mostly induce no or very mild symptoms in human. However, in some cases, EV infection can be associated with acute manifestations, such as hand, foot, and mouth disease (HFMD), pneumonia, myocarditis, aseptic meningitis or poliomyelitis-like acute flaccid paralysis. EV epidemics are detected in Europe on a regular basis, and often travel through different countries. The Austrian National Reference Centre (NRC) for Enteroviruses is responsible for PV surveillance and diagnosis of suspected EV cases. Their diagnosis methods are based on WHO recommendations, and cell line culture is used to isolate EV from stool or CSF samples. The presence of EV in the culture is assessed using RT-qPCR (targeting the highly conserved 5' non-coding region). For positive samples, EV are typed by sequencing of the VP1 region.

With the development of sequencing technology and the reduction of its cost, genome-wide analysis will progressively become the norm in diagnosis. In the case of EV, which evolve at a fast rate, sequencing-based diagnosis would be valuable as it would handle divergence better than other genotyping techniques. Whole genome sequencing (WGS) is also more informative than other typing techniques. It would allow more precise characterisation of the isolates and tracking of new recombination events.

To anticipate the progressive switch to more sequence-based diagnostic techniques, we implemented WGS based on a CDC protocol for E-30. This protocol included RNA extraction, complementary DNA synthesis, second strain DNA synthesis, cleaning, library preparation and Illumina sequencing. A bioinformatics pipeline inspired by metagenomics workflows was designed to identify EV species present in the sample. This workflow was compared with an online tool for metagenomics analysis. The whole procedure was tested for five EV species regularly identified at the NRC.

Role of the fellow: the fellow tested several protocol options and designed the final laboratory procedure. She also designed the bioinformatics tools to use for data analysis. She wrote a standard operating procedure and a simplified user guide for both the laboratory and bioinformatics steps.

3.2. Comparing nucleic-acid detection methods for SARS-CoV-2 diagnosis

Supervisor: Peter Hufnagl

SARS-CoV-2, the causing agent of COVID-19 in humans, has emerged in late 2019 in the province of Wuhan, China. The impact of COVID-19 in the different European countries varies greatly. Factors influencing the outcome of SARS-CoV-2 epidemic in each country are hard to assess, but some broadly accepted ones are early implementation of social distancing measures, sufficient hospital capacities and broad testing policies. SARS-CoV-2 testing has become a burning topic worldwide. At first, only major reference laboratories and hospitals were able to test for SARS-CoV-2, mostly using in-house RT-qPCR. Expanding the number of laboratories performing SARS-CoV-2 detection is essential to increase test volume, but smaller laboratories might not be adapted to these in-house techniques. In such facilities, commercial systems are more convenient. Many biotechnology companies have developed new SARS-CoV-2 detection kits, adapted to their existing system. But little data are available about these kits, and manufacturer's data are not always reliable. An independent testing of commercial SARS-CoV-2 detection kits would be much helpful for laboratories willing implement SARS-CoV-2 diagnostic, and further strengthen the national testing capacities.

We selected a panel of 93 samples from suspected COVID-19 patients, including 17 positives and 76 negatives. These samples were tested upon reception using AGES reference technique, Charité E-gene RT-qPCR (in-house). This panel was used to evaluate the following SARS-CoV-2 nucleic-acid based detection methods:

- Charité N-gene confirmatory RT-qPCR (in-house)
- Charité RdRp-gene confirmatory RT-qPCR (in-house)
- ViroReal Kin SARS-CoV-2 & SARS (Ingenetix)
- Isolthermal RT-qPCR (MIRAI GENOMICS)
- Detection kit for 2019 novel Coronavirus (2019-nCoV) RNA (DAAN)
- RealTime SARS-CoV-2 Assay for m2000 automated system (Abbott)
- VereCoV™ Detection Kit for VerePLEX™ Biosystem (Veredus)

The selected techniques were evaluated for reliability (discordant results), easy processing and interpretation, hands-on time and performance (samples per hour), quality (internal and external controls) and biosafety. Results of these evaluations were made public, so that laboratory interested in implementing these techniques would have reliable information on their advantages and weaknesses.

Role of the fellow: the fellow participated in the testing of some of the techniques involved in this comparison study (Charité RdRp-gene, DAAN detection kit, Abbott m2000 and VereCoV™ Detection Kit). She published the result of the VereCoV™ Detection Kit evaluation.

3.3. Training modules related to the assignment/projects

EPIET/EUPHEM Introductory Course: public health microbiology was covered during this module, together with its importance for surveillance and outbreak investigation. Classic laboratory methods and their interpretation were explained through lectures. Several case study included laboratory findings, which helped solving the outbreaks. One case study also focused on calculating specificity, sensitivity, positive predictive value and negative predictive value.

Outbreak investigation module: whole genome sequencing and its value for outbreak investigation in surveillance was covered in this module. It included a lecture on the various type of sequencing technique and an exercise on phylogenetic data interpretation.

Vaccinology module: a lecture about the different types of COVID-19 vaccines and how they induce immunity was delivered.

Educational outcome

The fellow learned to apply her technical microbiology knowledge to the high standards of diagnostics and public health laboratories. She also gained extensive knowledge in sequencing techniques, from sample preparation to data analysis.

4. Biorisk management

4.1. Implementing SARS-CoV-2 diagnosis by RT-qPCR

Supervisors: Daniela Schmid, Alexander Indra, Peter Hufnagl

On 31st of December 2019, Chinese authorities reported a cluster of pneumonia in the region of Wuhan. On the 9th of January 2020, the etiologic agent responsible for this epidemic has been identified as an unknown coronavirus. As early as January 2020, the Austrian health authorities started to be prepared a detailed preparedness plan, including diagnostic capacities.

The reference laboratory for Austrian SARS-CoV-2 diagnosis was the Virology department of Vienna Medical University. It was decided that AGES would be acting as a backup for SARS-CoV-2 suspected samples diagnosis. AGES molecular microbiology department was responsible for the implementation of SARS-CoV-2 diagnosis, the development of a standard procedure, including biosafety precautions, and the production of guidelines for sample collection and transportation.

Respiratory samples from suspected SARS-CoV-2 patients were handled in a BSL-2 laboratory, under a laminar flow and with additional PPE (disposable gown, FFP3 mask, first pair of long sleeve gloves and second pair of gloves). Samples were inactivated in order to be safely handled in BSL-1. The following steps of RNA extraction and RT-qPCR did not require specific biosafety precautions.

Extracted RNA was be tested for SARS-CoV-2 using two successive RT-qPCR: one targeting the E gene for screening (recognising SARS-CoV-2 but also SARS and MERS CoV), one targeting the RdRp gene for confirmation (specific for SARS-CoV-2). The limit of detection was 3.8 to 8.3 RNA copy/reaction depending on the target gene. Positive controls were ordered from the European Virus Archive. These controls consisted in RNA from Wuhan

coronavirus 2019 E gene and SARS-CoV Frankfurt 1. An internal control was included, to test for RNA extraction quality and PCR inhibition.

Role of the fellow: the fellow participated in the design of the test request form and the sampling/transportation instructions. Using WHO guidelines, she defined the biosafety precautions to be taken when handling SARS-CoV-2 suspected samples. She implemented and validated the whole SARS-CoV-2 diagnostic workflow. She wrote the standard operating procedure that was included in AGES quality management documents.

4.2. Training modules related to the assignment/projects

The fellow already had extensive experience in this area. She worked in BSL-2, BSL-3 and BSL-4 laboratories before starting her fellowship.

Biorisk and Quality Management module: this planned module was cancelled due to the COVID-19 pandemic.

Educational outcome

The fellow's previous knowledge in biosafety was of importance at the beginning of the COVID-19 pandemic. With the help of colleagues, she defined the precautions to be taken when handling SARS-CoV-2 suspected samples. She also worked on sample collection and transportation guidelines.

5. Quality management

5.1. SARS-CoV-2 external quality assessment

Supervisors: Alexander Indra, Peter Hufnagl

In March 2020, all laboratory performing SARS-CoV-2 molecular diagnosis in Vienna, including AGES, were invited to participate to an External Quality Assessment (EQA). This EQA was organised by the national reference centre (NRC) for respiratory viruses (Center for virology, Vienna Medical University). The aim of the EQA was the comparison of RNA extraction and RT-qPCR used by various laboratories and the establishment of a detection limit for each protocol.

Five randomised respiratory samples from COVID-19 suspected patients were sent by the Austrian NRC for respiratory viruses to all facilities in Vienna performing SARS-CoV-2 diagnosis. Samples were be processed using routine diagnosis procedures in all facilities. Results were be sent to the NRC for comparison. Results of the comparison were be made anonymous and sent to all participants.

Role of the fellow: the fellow processed the EQA samples, wrote the EQA report for AGES and shared it with the EQA organisers.

5.2. Training modules related to the assignment/projects

Biorisk and Quality Management module: this planned module was cancelled due to the COVID-19 pandemic.

Educational outcome

The fellow learned and applied the quality standards necessary to high quality diagnostics. She developed a good understanding of quality control process. She was involved in an EQA on behalf of her training site.

6. Teaching and pedagogy

6.1. Blood parasite diagnosis by microscopy

Supervisor: Franz Allerberger

AGES, Vienna Veterinary University and ÖGTPM (Austrian Society for Tropical Medicine, Parasitology and Migration Medicine) organised in November 2019 a one-day workshop entitled 'Hands-on-Workshop: Update Microscopy'. This workshop included both theoretical and practical session and covered stool parasites, blood parasites, *Borrelia*, *Rickettsiales* and *Leptospira*. The fellow participated in the practical session of 'Blood parasite identification by microscopy'. Eighteen participants, all from the medical or biology field, attended the workshop. The purpose was to familiarise healthcare workers with the identification of parasites by microscopy. As most parasitic diseases are still rare in Austria, sensitisation of health care worker to their diagnosis would improve and accelerate their detection.

6.2. Case study facilitation: Emergence of COVID-19 in a District in Myland

Supervisor: Peter Kreidl

The fellow was invited to facilitate a case study at the Medical University of Innsbruck in October 2020. The case study entitled 'Emergence of COVID-19 in a District in Myland' was developed Peter Kreidl, who also organised the case study sessions. Participants were seventh semester medical students, and this case study was included in their first public health and epidemiology course. Each group of five to seven participants had two sessions of one and a half hours with a facilitator to go through the case study. The fellow helped review the case study material, facilitated two groups and developed a post-course survey.

6.3. Online seminar: Ebola, the facts behind the myth

Supervisor: Werner Ruppitsch, Alexander Indra

The fellow gave a lecture in an online seminar organised by AGES Akademie in January 2021. The AGES Akademie seminars are organised once a month as part of the AGES continuous training plan. The programme is in line with the AGES basic mission for health and food safety. It is open to AGES employees, but also other health professionals. Participants are granted credentials that are acknowledged by two continuous education programs: the Medical Association of Vienna (for medical doctors) and Biomed Austria (for biomedical analysts).
The fellow chose Ebola as the seminar topic. The purpose was to entangle the rumours surrounding Ebola. This online seminar went back to the basis of Ebola virus, its emergence, its transmission, and its pathogenicity. From these facts, it constructed a reliable picture of what Ebola is, and how to efficiently face it.

6.4. Online case study facilitation: An outbreak of gastro-enteritis in Kalundborg, Denmark

Supervisor: Pieter-Jan Ceyssens

In March 2021, the fellow was invited to facilitate an online case study in a course organised by Natalie Fischer (EUPHEM fellow) and Pieter-Jan Ceyssens (EUPHEM supervisor), from Sciensano, Belgium. This course was included in a module about possible career perspectives for bioengineering students of the Katholieke Universiteit Leuven, Belgium. It consisted in a presentation of Belgium public health system and the role of Sciensano (by Pieter-Jan Ceyssens), followed by a case study. The case study was adapted from the ECDC case study 'An outbreak of gastro-enteritis in Kalundborg, Denmark' and facilitated by Natalie Fischer, Pieter-Jan Ceyssens, Andreas Hoefer (EUPHEM fellow, Spain), Justine Schaeffer (EUPHEM fellow, Austria) and Heloise Lucaccioni (EPIET fellow, Portugal).

6.5. Online case study facilitation: Multidrug resistant *Salmonella* Kentucky scenario exercise

Supervisor: Aftab Jasir

The third part of the EPIET/EUPHEM Introductory Course for cohort 2020 included a whole genome sequencing (WGS) themed case study. This case study consisted in the analysis of an outbreak of multidrug resistant *Salmonella enterica* serotype Kentucky outbreak using sequencing data. The fellow facilitated a group of 6 participants (5 EPIET fellows and 1 EUPHEM fellows) with Natalie Fischer (EUPHEM fellow) in April 2021. The objective of this case study were to provide fellow with background knowledge on WGS, have them perform WGS analyses with open-source tools and illustrate the benefit of genetic and genomic data for outbreak investigation.

6.6. Pilot testing of an innovative teaching method: the Flu-Game

Supervisor: Peter Kreidl

'Influencing Influenza' is an interactive game developed by Transmissible and Jade Owl Studios for the University of Utrecht (Netherlands). This game was designed for medical students to better understand the roles of relevant organisations in outbreak response and the challenges of such response (time pressure, limited resources …). The Medical University of Innsbruck adapted this game to Austria, by translating it to German and adjusting the relevant organisations. They plan to use this game as a practical course for seventh to eighth semester medical students. The fellow was invited to join the pilot-testing of the Austrian version among 10 participants. For the pilot session, two group of five participants played the Austrian version of the game under the supervision of the organisers. It was by a hot debrief and the filling of a feedback survey.

6.7. Training modules related to the assignment/projects

EPIET/EUPHEM Introductory Course: a teaching workshop was organised during this module. It focused on the specificities of adult learning, the importance of engaging the audience and alternate teaching formats.

Management, Leadership and Communication in Public Health module: communication tools that can be easily adapted to teaching were provided during this module. It included basic knowledge on the different communication and learning styles, but also practice sessions.

Educational outcome

During her fellowship, the fellow got her first real teaching experience. She gained increasing levels of responsibilities, from support during the course to working on learning material. She taught different types of audience which required specific adaptations of the course content and design.

7. Public health microbiology management

7.1. Project management

Supervisors: all project supervisors

The fellow was principal investigator or involved in project management for most of her projects, which covered various aspects of public health microbiology management. Corresponding duties included project development and timeline definition, work in multidisciplinary teams, collaboration with AGES and non-AGES partners and findings communication.

7.2. Recruiting stakeholders for a Hantavirus surveillance system

Supervisors: Georg Duscher, Franz Allerberger

Hantavirus surveillance is an increasingly important topic for Austrian public health policies. The setting up of a surveillance system in rodents fitted in the missions of AGES, as defined by the Austrian Ministry of Health. However, such a system required the participation of several internal and external stakeholders. The fellow participated in the recruitment the stakeholders, under the supervision of her project supervisors. She worked on how to best organise the system to minimise the additional workload for each stakeholder and the risk of errors. She designed documents for participant information and consent, sample follow-up and result communication.

7.3. COVID-19 management in Austria

Supervisors: Daniela Schmid, Alexander Indra, Peter Hufnagl

From January 2020 onwards, the fellow was involved in COVID-19 pandemic preparedness and management. She attended several meetings coordinating case management, surveillance, and diagnostic efforts in the province of Vienna. She implemented SARS-CoV-2 diagnostic at AGES and participated in the constant optimisation of the workflow. She tested many different SARS-CoV-2 diagnostic techniques, developed by private companies or public research laboratories, and emitted independent opinions on the advantages and weaknesses of each techniques. Such independent evaluations were communicated to Austrian laboratories.

The fellow had a particularly fruitful collaboration with the Viennese Institute of Molecular Biotechnology (IMBA), which developed two SARS-CoV-2 diagnostic techniques. One was a LAMP-PCR, which had the advantages to require very little equipment and reagents. The second was sequencing based, and allowed the testing of tens of thousands of samples at the same time for SARS-CoV-2 and other common respiratory viruses. The fellow was trained to both techniques and assisted the IMBA teams with their validations.

7.4. International Assignment: COVID-19 sero-epidemiological study in Lithuania

Supervisors: Erika Duffell, Maria Keramarou

In May 2020, the fellow was selected with Giuseppina Ortu (EPIET fellow, France) for an ECDC-coordinated international assignment. The role of the two fellows as defined in the initial project was:

- Support the local team in adapting the WHO study protocol to the local setting;
- Provide ongoing epidemiological support to the local team in implementing the protocol;
- Provide support to the local team in choosing the appropriate antibody testing method for the needs of the study and the provide support for the validation of the test method, prior to use in the field
- Support the analysis of the results of the survey and draft a report and a paper for peer review publication based on the findings

The fellows and supervisors had a first meeting with the Lithuanian officials in charge of the study. During this meeting, ECDC offered support from the fellows on any area where needed. This offer was finally declined by the Lithuanian officials.

Since then, the fellow has been attending joint ECDC and WHO Europe online meeting on COVID-19 sero-epidemiological studies, organised once a month.

7.5. Cross-border outbreak of Listeria monocytogenes Ny9

Supervisors: Ariane Pietzka, Franz Allerberger

Two Austrian listeriosis cases were detected in November 2020. The fellow was involved in the outbreak investigation. Sequencing of the *Listeria monocytogenes* clinical isolates showed that the Austrian cases belonged to a Europe-wide outbreak. Following this finding, a close collaboration begin with the outbreak investigation teams of other affected countries. It was a good example of successful cross-border data sharing and resource combination. Epidemiological data, investigation hypothesis and isolate sequences were largely shared through the European listeriosis network. This efficient collaboration allowed a quick resolution of the outbreak on the Austrian side.

7.6. Training modules related to the assignment/projects

Management, Leadership and Communication in Public Health module: a large part of this module focused on management and leadership. The organisation of public health at EU level was presented. Management concepts, such as John Adair's management model, the wheel of success, management styles and team dynamics, were taught and experimented by the fellows. A full day was dedicated to crisis management and working under pressure.

Vaccinology module: the specificity of management and communication of vaccine-preventable diseases were covered by this module. The epidemiological and political aspects to consider before implementing a new vaccine were also discussed.

Educational outcome

As some of the fellow's project involved an important number of actors, the fellow gained experience in managing teams with various backgrounds. She discovered the complexity of public health networks at national and international levels, and learned how to navigate them. She also applied some of the techniques learned to her independent work, in order to increase her efficiency.

8. Communication

8.1. Publications related to the EUPHEM fellowship

J. Schaeffer, W. Ruppitsch, **Evaluierung eines "lab-on-a-chip"-Nukleinsäure-Nachweissystems SARS-COV-2: VerePLEX™-Biosystem**, AGES – *Wissen aktuell* May 2020 (https://www.ages.at/en/wissen-aktuell/publikationen/evaluierung-eines-lab-on-a-chip-nukleinsaeure-nachweissystems-sars-cov-2-vereplextm-biosystem/)

J. Schaeffer, S. Huhulescu, A. Stoeger, F. Allerberger, W. Ruppitsch, **Draft genome sequences of six Corynebacterium ulcerans strains isolated from humans and animals in Austria, 2013 to 2019**, *Microbiology Resource Announcements* Sep 2020, 9 (36) e00946-20 (doi: 10.1128/MRA.00946-20)

J. Schaeffer, S. Huhulescu, A. Stoeger, F. Allerberger, W. Ruppitsch, **Assessing the genetic diversity of Austrian Corynebacterium diphtheriae clinical isolates, 2011-2019**, *Journal of Clinical Microbiology* Dec 2020, JCM.02529-20 (doi: 10.1128/JCM.02529-20)

J. Schaeffer, S. Revilla-Fernández, E. Hofer, R. Posch, A.Stoeger, C. Leth, F. Schmoll, V. Djordjevic, B. Lakicevic, K. Matovic, P. Hufnagl, A. Indra, F. Allerberger, W. Ruppitsch, **Tracking the Origin of Austrian Human Brucellosis Cases Using Whole Genome Sequencing**, *Frontiers in Medicine* Feb 2021, Fmed.635547 (doi: 10.3389/fmed.2021.635547)

R. Yelagandula, A. Bykov, A. Vogt, R. Heinen, E. Özkan, M. Strobl, J. Baar, K. Uzunova, B. Hajdusits, D. Kordic, E. Suljic, A. Kurtovic-Kozaric, S. Izetbegovic, J. Schaeffer, P. Hufnagl, A. Zoufaly, T. Seitz, VCDI Consortium, M. Födinger, F. Allerberger, A. Stark, L. Cochella, and U. Elling, **Multiplexed detection of SARS-CoV-2 and other respiratory infections in high throughput by SARSeq,** *Nature Communications* March 2021, NCOMMS-20-47402B (doi: 10.1038/s41467-021-22664-5)

J. Schaeffer, S. Revilla-Fernández, E. Hofer, V. Djordjevic, B. Lakicevic, A. Indra, F. Schmoll, F. Allerberger, W. Ruppitsch, **Tracking brucellosis – a re-emerging disease**, IOP Conference Series: Earth and Environmental Science, July 2021 (submission for the 61st International Meat Industry Conference call for papers)

8.2. Reports

Standard Operating Procedure: SARS-CoV-2 diagnostic by RT-qPCR, completed in March 2020

External Quality Assessment report: molecular detection of SARS-CoV-2 nucleic acid, completed in May 2020

Standard Operating Procedure: molecular detection of SARS-CoV-2 nucleic acid, completed in May 2020

Outbreak Investigation report: Coronavirus pandemic – overseeing AGES coronavirus diagnosis, completed in June 2020

Study protocol: molecular surveillance of Hantaviruses in rodent surrounding the homes of patients infected by Hantaviruses, completed in February 2021

International Assignment report: Technical support to Lithuania to conduct a sero-epidemiology study for COVID-19, completed in September 2020

Technique development report: adapting and implementing a single nucleotide polymorphism-based typing method to support *Mycobacterium avium* subspecies *paratuberculosis* surveillance, completed in November 2020

Study protocol: assessing genetic determinants of rifampicin resistance carried by the *rpoB* gene in Austrian *N. gonorrhoeae* isolates, completed in December 2020

Outbreak Investigation report: Cross-border outbreak of *Listeria monocytogenes* Ny9, completed in January 2021

Standard Operating Procedure: Hantavirus detection in rodent organs using pan-Hantavirus nested PCR, completed in February 2021

Report on the implementation of a surveillance system: molecular surveillance of Hantaviruses in rodent surrounding the homes of patients infected by Hantaviruses, completed in February 2021

Standard Operating Procedure: whole genome sequencing of Enteroviruses, completed in March 2021

8.3. Conference presentations

Tracking the origin of Austrian human Brucellosis cases using whole genome sequencing, 2005-2019, J. Schaeffer, S. Revilla-Fernandez, E. Hofer, A. Stoeger, C. Leth, F. Schmoll, B. Lakicevic, A. Indra, F. Allerberger, W. Ruppitsch, pre-recorded presentation and moderated fireside panel, ESCAIDE – Food and waterborne diseases outbreaks session (1b), November 2020

Rifampicin resistance associated with *rpoB* mutations in *Neisseria gonorrhoeae* strains isolated in Austria, 2017-2020, J. Schaeffer, K. Lippert, S. Pleininger, A. Stöger, P. Hasenberger, S. Stadlbauer, F. Heger, A. Eigentler, A. Geusau, A. Indra, F. Allerberger, W. Ruppitsch, ePoster, ECCMID – Resistance surveillance & epidemiology: Gram-negatives session (3b), July 2021

Genomic characterisation of high-level macrolide resistance in *Campylobacter* isolates in Austria, 2016-2020, J. Schaeffer, N. Hiden, A. Murer, W. Ruppitsch, S. Koeberl-Jelovcan, ePoster, ECCMID – Resistance surveillance & epidemiology: Gram-negatives session (3b), July 2021

Phylogenomic relatedness between Austrian *N. gonorrhoeae* strains correlates with antimicrobial resistance patterns, J. Schaeffer, K. Lippert, S. Pleininger, A. Stöger, P. Hasenberger, S. Stadlbauer, F. Heger, A. Eigentler, A. Geusau, A. Indra, F. Allerberger, W. Ruppitsch, ePoster, ESCAIDE – Microbiology session, November 2021

8.4. Other presentations

Kick-off meeting for EUPHEM: presentation to introduce the fellow's background to potential project supervisors and discuss EUPHEM projects, 30/01/2020 (2h)

Communication to a higher authority: presentation on behalf of the working group (two EUPHEM fellows from C2018 and two EUPHEM fellows from C2019) on the future of the fellowship program, 13/02/2020 (10 min presentation, 10min questions)

Seminar at the Medical University of Innsbruck: Highly pathogenic viruses and where to find them, 16/10/2020 (40 min presentation, 20min questions)

AGES Akademie online seminar: Ebola, the facts behind the myth, 11/01/2021 (30 min presentation, 20min questions)

8.5. Other activities

Footage showing how AGES handles suspected 2019-nCoV samples, May 2020
This 2min video shows every step of sample processing, from reception to result analysis. It emphasised biosafety precautions.

Correcting COVID-19 themed document for children's English courses, level A2-B1 (http://www.FOR-EVERy1.com), June 2020
FOR-EVERy1 developed some materials based on the COVID-19 pandemic and how it affects school-aged children. They asked AGES to proof-check the materials, in order to avoid spreading misinformation.

Review of scientific papers for peer-reviewed journals
- Two manuscripts on SARC-CoV-2 diagnostic methods for Clinical Microbiology and Infection
- Three manuscripts on SARS-CoV-2 immunity (x2) and BCG vaccine for Journal of Applied Microbiology
- One manuscript on *Salmonella* genomic surveillance for Epidemiology and Infection

8.6. Other training modules

EPIET/EUPHEM Introductory Course: theoretical knowledge on communication was provided during this module, for example the structure of written communication, the definition of the target audience, or formulating recommendations. Fellows made a three-minute presentation on a topic of their choice and gave constructive feedback. The research project was also presented to other fellows and coordinators/facilitators.

Management, Leadership and Communication in Public Health module: tools for efficient communication were given during this module. They included basic knowledge on the different communication and learning styles, but also practice sessions. The fellow was in charge of presenting her group project for the 'communication to a higher authority' exercise.

Rapid Assessment and Survey Methods module: a lecture on risk and crisis communication was delivered during this module. A case study was focusing on the communication aspects of outbreak investigation.

Project Review modules: during the two project review modules, fellows prepared and delivered an oral presentation on one of their projects. Extensive feedback was given by other fellows, regarding the content but also its delivery.

9. EPIET/EUPHEM modules attended

- **Introductory Course (IC)**, Spetses, Greece, from 23 September to 11 October 2019
- **Outbreak Investigation Module**, Ministry of Health Cyprus and the European University Cyprus, Nicosia, Cyprus, from 4 December to 8 December 2019
- **Management, Leadership and Communication in Public Health Module**, ECDC, Stockholm, Sweden, from 10 February to 14 February 2020
- **Multivariable Analysis Module**, organised online by the Robert Koch Institute, from 20 April to 24 April 2020
- **Project Review Module**, organised online by ECDC, from 24 August to 27 August 2020
- **Time Series Analysis Module**, organised online by ECDC, from 25 January to 29 January 2021
- **Rapid Risk Assessment**, organised online by ECDC, 27 April, 4 May and 5 May 2021
- **Vaccinology Module**, organised online by ECDC, from 14 June to 18 June 2021
- **Project Review Module**, organised online by ECDC, from 24 August to 27 August 2021

10. Other training

- **German language courses A1 to B1**, seven modules of 40 hours, Internationales Kulturinstitut, Vienna, Austria
- **Whole genome sequencing platform**, technical training, AGES, October 2019 (three days)
- **Attending the European Scientific Conference on Applied Infectious Disease Epidemiology (ESCAIDE)**, Stockholm, Sweden, from 27 November to 29 November 2019 (three days)
- **Emerging respiratory viruses, including nCoV: methods for detection, prevention, response and control**, OpenWHO online course, followed in January 2020 (2h)
- **MOOC: Investigate Health, how does it work?**, online course (in French) on how to design and implement a questionnaire study for health-related topics, followed in February-March 2020 (around two hours per week for six weeks)
- **Briefing & Exchanges on China's COVID-19 Experiences with Leading Experts**, webinar, Dr. Zhong Nanshan, Dr. Li Lanjuan, Dr. Qiao Jie, Dr. Zhu Chouwen, Dr. Zhang Junhua, Dr. Paul Rothman, Dr. Davidson Hamer, Dr. Ian Lipkin, 19 March 2020 (2h30)

- **ESCMID eAcademy webinars on COVID-19**
  Infection Prevention & Control in the Time of COVID-19, 27 April 2020 (3h)
  COVID-19 diagnostics: Lights and shadows – 7 May 2020, 07/05/2020 (3h)
- **Training on investigation of fungal infection**, Division of Hygiene and Medical Microbiology at the Medical University of Innsbruck, Austria, from 12 October to 15 October (four sessions of two to three hours)
- **Attending the European Scientific Conference on Applied Infectious Disease Epidemiology (ESCAIDE)**, online, from 24 November to 26 November 2020 (three days)
- **Legal Epidemiology and the Path to Better Health Law and Policy**, webinars organised by UK Faculty of Public Health and the European Public Health Association, 21 January 2021 (1h30)
- **2021 ISV Virtual Congress -COVID-19 Vaccine Update**, webinar organised by the International Society for Vaccines, 10 February 2021 (3h)
- **Disinformation, Conspiracy Theories, and COVID-19 Vaccines**, webinar organised by WHO EURO, 19 February 2021 (1h)
- **Risk communication during the COVID-19 pandemic**, webinar organised by the EPIET Alumni Network, 2 March 2021 (1h)
- **Go.Data in the times of COVD-19 seminar series**, webinar organised by the EPIET Alumni Network, 30 March 2021 (1h)
- **Setting up and running a genetic sequencing facility**, WHO Public Health Laboratories Webinars series, 21 April 2021 (1h30)
- **Global consultation on SARS-CoV-2 variants of concern and their impact in public health interventions**, WHO webinar, 10 June 2021 (3h30)
- **Webinar on Genomic Surveillance and National Public Health Institutes**, webinar organised by the IANPHI, 30 June 2021 (2h)
- **Attending the 31st European Congress of Clinical Microbiology & Infectious Diseases (ECCMID)**, online, from 9 July to 12 July 2021 (four days)
- **Webinar on Implementation of SARS-CoV-2 antigen-RDTs**, WHO webinar, 14 July 2021 (1h30)

Discussion

Coordinator's conclusions

One of the main goals of the EUPHEM programme is to expose fellows to diverse and multidisciplinary public health experiences and activities, thus enabling them to work across different disciplines. This report summarises all activities and projects conducted by Justine Schaeffer during her two-year EUPHEM fellowship (cohort 2019) as an EU-track fellow at AGES (Austria). As a young and talented virologist, Justine spent her two years of Fellowship being involved in a variety of projects spanning many different disease groups: among others, *Listeria monocytogenes*, *Corynebacterium*, *Brucella melitensis*, *Campylobacter*, *Neisseria gonorrhoeae*, Hantavirus, Enterovirus and, last but not least, SARS-CoV-2, for which she has actively contributed both in terms of outbreak investigation and laboratory diagnosis. Justine has been instrumental in setting up projects exploring the beneficial use of whole genome sequencing in the public health setting and her success is demonstrated by the amazing amount of work done and outputs produced, as testified by this portfolio. Despite her young age she is an accomplished professional who took her role as a fellow seriously and took every opportunity to expand her knowledge and her experience. Being her frontline coordinator has been a pleasure and I, together with the EUPHEM team, wish her the best in her future, undoubtably brilliant, career in public health microbiology.

Supervisor's conclusions

Justine impressed with her interest, accurate, focussed and speedy work style. She managed all tasks and projects during her fellowship so fantastic and professional that supervision and working with her were simply a great pleasure and honour for me. As a virologist she became familiar with new methods (WGS) and bacterial pathogens very quickly, which finally resulted in excellent publications. Among the excellent results and outputs, I would like to highlight her leading role at AGES in establishing SARS-CoV-2 testing. Without her knowledge and expertise in virology, AGES wouldn't have been able to establish SARS-CoV-2 testing so straightforwardly and rapidly. Justine was a very enjoyable colleague and established rapport very easily with all group members. As a result, she immediately became an indispensable member of our group. Justine is an outstanding expert and, although it saddens me that she will leave, I am happy that she found a position in the field of public health in France and I wish her all the best and success for this new job and career.

Personal conclusions of fellow

The EUPHEM fellowship has been a life-changing experience for me. It gave me an opportunity to re-invest my academic background in the field of public health. I gained an extensive knowledge on how national and international public health systems work. I learned the fundamental concepts of epidemiology and the analytical skills of this new discipline. I also met fascinating people from very different areas and hope these relationships will continue to grow in the future.

I could not have hoped for a better training site than AGES. I felt trusted and valued as a public health professional. I acquired a strong expertise in sequencing and applied it to various pathogens of public health importance. The COVID-19 pandemic was a challenging time, but also an opportunity to support AGES using my virology background. I got involved both inside and outside the laboratory, which was a rich experience for me and, I hope, a benefit for my training site.

There are things that I wished went differently, but that is always the case when you go through such an intense experience. Everything I learned during these two years is highly valuable for my career. Having the opportunity to work on so many different projects in parallel fitted me perfectly, and I hope that my future will be as stimulating.

Acknowledgements of fellow

I will be forever grateful to my supervisor, Werner Ruppitsch. He provided me with diverse and engaging projects, the freedom to manage them as I saw fit, and unconditional support whenever I needed it. I also want to thank all my project supervisors, with whom it was a pleasure to work. A sincere thanks also to my other colleagues who have always been supportive and helpful, no matter how much I butchered their language. A special thanks to Lukasz Henszel, EPIET fellow 2018, for the lunchtime psychology session and the guidance through the beginning of my fellowship. Finally, a heartfelt thanks to Adriana Cabal, who not only pilot-tested the EUPHEM fellowship at AGES (allowing it to become such a good training site for me) but was also an immeasurable support and a great friend throughout these two years.

I would also like to thank ECDC for letting me into this program, and for having such a program in the first place. I also thank the faculty office and the module organisers, and I know the huge amount of work they are doing for us. I thank my frontline coordinator for guiding me through the fellowship requirements and the other fellowship coordinators who provided me with additional input during my midterm review. I also thank the EAN network who organised various side events that I really enjoyed and the members I was lucky enough to meet.

Last but not least, I thank my fellow EPIET, EUPHEM and PAE for entering this program and giving me a chance to meet them. This includes C2018 and C2020 fellows I was able to meet, but most of all my friends from C2019. I am immensely grateful to have met you all, and wish you the best for the future. C2019 is and will remain the best cohort of all time.