

FINAL REPORT

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European Public Health Microbiology Training Programme (EUPHEM), 2021 Cohort
Greek EUPHEM Consortium, Greece

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

Ioanna Spiliopoulou is a Medical Doctor and a state accredited Medical Biopathologist (2007). She completed her PhD thesis in the Faculty of Medicine, National and Kapodistrian University, Athens, Greece and graduated in 2013 (Grade 'Excellent'). She received a Master of Science's degree in public health (specialisation: infectious diseases) from the National School of Public Health, Athens, Greece in 2017 (Grade 'Excellent'). Since 2007, Ioanna has been working at the Central Public Health Laboratory (CPHL) of the National Public Health Organization at the Water Microbiology Laboratory, and became head of the laboratory in 2009. Ioanna is also deputy head of the Food and Bottled Water Microbiology Laboratory and was technical manager for microbiological methods of CPHL (according to ISO 17025) from 2016 until the start of the fellowship. She has participated as a substitute member in the working group for implementing the new EU Drinking Water Directive in the national legislation and in the subcommittee for the recognition of waters as natural mineral waters. Her work experience has mainly dealt with food- and waterborne diseases. To gain deeper knowledge and broaden her perspectives in public health microbiology through the development of core competencies specific to disciplines she wasn't familiar with, she applied for the ECDC Fellowship on European Public Health Microbiology Training Programme. In this way she could serve better the public health microbiology community at both national and EU levels.

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 EPIET core competencies, as set out in the ECDC Fellowship Manual¹.

1. Epidemiological investigations

1.1. Outbreak investigations

A community waterborne outbreak due to Salmonella Bovismorbificans, Greece, August 2022

Supervisors: Dr Kassiani Mellou, Mrs Theologia Sideroglou and Assist. Prof. Georgia Mandilara

Category: *Food- and waterborne diseases*

In August 2022, the Greek National Public Health Organisation was notified about increased gastroenteritis cases in a small town (A) within the Regional Unit of Argolida, Peloponnese Region, southern Greece. We conducted an outbreak investigation to identify the source and implement control measures. In an unmatched 1:3 case-control study, cases and controls were defined as people residing or visiting town A between 10 and 24 August 2022, with and without diarrhoea, vomiting, and fever, respectively. They were interviewed about food and drink exposures. Cases' stool samples were cultured on agar plates and further characterised by serotyping, antimicrobial susceptibility testing (Kirby-Bauer method) and Pulse Field Gel Electrophoresis (PFGE). Environmental investigations included tap water sampling for microbiological and chemical analysis in town A, and inspection of the water supply system. We identified 33 cases (55% female) with a median age of nine years (range: 0–79 years). Tap water consumption was the only significant risk factor (OR=5.46, 95%CI=1.02-53.95). *Salmonella enterica enterica* ser. Bovismorbificans (6,8:r:1,5) isolated from stool (n=8) and a tap water sample (n=1) had identical PFGE profiles. No resistant isolates were identified. *Salmonella enterica enterica* ser. Bardo (8:e,h:1,2) was detected in the water supply system, and chlorination levels were found to be below recommended limits in samples prior and during the outbreak. Despite investigations, no conclusion was drawn on how bacterial contamination occurred. We released public advice to consume bottled water and to adhere to strict hand hygiene rules until tap water was declared safe for drinking. We also recommended regional and municipal water safety authorities to monitor more efficiently the water supply systems, towards water safety planning, to mitigate risks. Epidemiological, laboratory and environmental investigation revealed a waterborne *Salmonella Bovismorbificans* outbreak in town A in August 2022, which lasted 15 days as no new cases were reported after 24 August 2022.

The fellow participated in the different stages of the outbreak investigation as a co-investigator. She conducted telephone interviews with patients and controls, performed statistical analyses with the use of Stata and contributed to data interpretation in collaboration with the Greek EPIET fellow Lida Politi. She participated in all the

¹ 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>

steps of the laboratory investigation: serotyping of *Salmonella* strains referred to the National Salmonella Shigella Reference Centre, antimicrobial resistance of referred strains using the Kirby-Bauer method, molecular typing by the DNA digestion method with restriction endonuclease and electrophoresis in a pulsed field (PFGE) and saw how the dendrogram was constructed with the Bionumerics Program 6.6. She also wrote the outbreak report (see 8.1.2.1) and contributed to writing two abstracts accepted as an oral presentation at the 13th Panhellenic Conference of the Panhellenic Association of Bioscientists 2022 (see 8.3.4) and as a poster presentation at European Scientific Conference on Applied Infectious Disease Epidemiology (ESCAIDE) 2023 (see 8.2.4).

Educational outcome

The fellow participated in a multidisciplinary outbreak control team and had hands-on involvement in outbreak investigation; active case finding, data collection, data analysis, generating and testing appropriate hypotheses, hands-on laboratory analysis of referred strains, laboratory results interpretation and communication, and formulation of recommendations. The importance of non-sequencing typing method in confirming the outbreak and its significance in adapting the case definition during the outbreak investigation was consolidated. Additionally, the fellow communicated results in various ways such as writing an outbreak report and abstracts in national and international conferences.

1.2. Surveillance

Evaluation and further analysis of genotyping *P. vivax* samples from malaria cases recorded in Greece during 2015-2019

Supervisors: Assoc. Prof. Eleni Patsoula, Mrs Anastasia Mpimpa, Dr Danai Pervanidou

During 2015-2019, 405 malaria cases were reported to the Hellenic National Public Health Organization; 372 were imported and 33 were locally acquired/introduced cases of which 32 (97%) were *P. vivax* malaria cases. The use of molecular tools that genetically fingerprint *P. vivax* parasites could provide a powerful tool as an adjunct to classical epidemiological investigations, especially in cases where medical or travel history is scarce, improving epidemiological case classification and contributing to the identification of unsuspected additional foci of local transmission. We studied 124 *P. vivax*-infected archived human blood samples, referred to the National Malaria Reference Laboratory (NMRL) from all over Greece during 2015-2019. The *P. vivax* populations were subjected to genotyping targeting 9 main polymorphic markers: the *Plasmodium vivax* merozoite surface protein-3a (*Pvmsp-3a*) locus, using a combined polymerase chain reaction/restriction fragment length polymorphism (PCR/RFLP) protocol, and 8 microsatellite loci (Pv1.501, Pv3.502, MS1, MS5, MS7, MS8, MS12 and MS20) using a semi-nested PCR and fragment analysis protocol. *P. vivax* isolates were assigned to specific haplotypes and the results were assessed for the existence of same or closely related haplotypes in the same regional unit the locally acquired *P. vivax* malaria cases were notified, during the same year of notification as well as during consecutive years. The genotypic analysis confirmed the epidemiological classification of notified cases as locally acquired *P. vivax* malaria cases and no additional not previously suspected foci of local transmission were identified. In only one cluster the index case could be identified. The risk of re-introduction of malaria in specific vulnerable and receptive rural Greek areas exists, especially where the presence of adequate numbers of competent vectors is combined with the presence of malaria patients coming from endemic countries. Molecular genotyping of the locally acquired cases is considered useful for future comparisons and will help guide timely and effectively interventions and control measures implementations.

The fellow performed literature review, familiarised herself with molecular-based (PCR, nested PCR) and genotypic techniques (fragment analysis), performed data cleaning, analysis, and evaluation of the laboratory and epidemiological data. Furthermore, the results of this project were presented orally during the Project Review Module 2023 (see 8.3.6) and a manuscript for a peer-reviewed scientific journal is in final preparation for submission (see 8. Communication/publications in preparation).

Educational outcome

The fellow was exposed for the first time to the concept of microsatellites as genetic markers and to the fragment analysis molecular technique used for genotyping. Additionally, during her training in the NMRL, she familiarised with light microscopic examination of a blood film, rapid diagnostic test (RDT) and PCR analysis of blood samples referred to the NMRL for verification of malaria diagnosis. She had also the opportunity to microscopically examine a bank of archived blood samples positive for all malaria species (*P. falciparum*, *P. vivax*, *P. ovale* & *P. malariae*). Microscopic examination of blood films and malaria parasite counting in received blood samples under the supervision of the experienced laboratory staff was also undertaken. Overall, the fellow familiarised herself with the applied concepts of parasitology to the public health disciplines; identified the use and limitation of diagnostic and typing methods and their interpretation in patient diagnosis, outbreak investigations, surveillance and epidemiological studies; acknowledged the need to integrate microbiological and epidemiological data in disease surveillance, and acquired experience in writing project proposal and a manuscript for a peer-reviewed scientific journal.

2. Applied public health research

Neisseria meningitidis: Characterisation of antibiotic resistance genes during 2010–2021 in Greece

Supervisors: Prof. Georgina Tzanakaki, Dr Athanasia Xirogianni.

For effective case management and chemoprophylaxis of Invasive Meningococcal Disease, prompt and appropriate antibiotic treatment is required. *N. meningitidis* is usually susceptible to antibiotics; however, reduced susceptibility to penicillin, ciprofloxacin, and rifampicin is increasing worldwide, jeopardising patients' outcomes. We assessed phenotypically and for the first time genotypically the antimicrobial resistance patterns of 192 strains isolated from IMD cases from all over Greece during 2010–2021. Serogroups were identified by slide agglutination test. All isolates were genotyped by Multilocus Sequence Typing (MLST). Minimum Inhibitory Concentration (MIC) to penicillin, rifampicin and ciprofloxacin was carried out by E-test and results were interpreted according to EUCAST guidelines. Further, *penA*, *rpoB* and *gyrA* genes were amplified by PCR and sequenced. Of the 192 isolates, 52% (99/192) were penicillin-susceptible/standard exposure (Pen^S), 37% (72/192) were penicillin-susceptible/increased exposure (Pen^I) and 11% (21/192) were penicillin-resistant (Pen^R). A reduction on the proportion of Pen^S isolates was observed ranging from 73% (2010) to 27% (2019). Almost all Pen^R isolates belonged to MenB (95%; 20/21) while 5% (1/21) belonged to MenW. Forty *penA* alleles were identified; *penA14*, *penA25* and *penA22* were related to reduced susceptibility to penicillin, while *penA9*, *penA910* and *penA295* to penicillin resistance. Two ciprofloxacin-resistant isolates harboured the *gyrA346* allele found for the first time in Greece during an outbreak in a migration camp, while one rifampicin-resistant isolate harboured the *rpoB5* allele. Resistance to ciprofloxacin and rifampicin remained rare. However, decreasing susceptibility was observed for penicillin. As Greece is one of the countries with high antimicrobial resistance, phenotypic antimicrobial resistance surveys of isolates collected from Greek IMD cases, along with genetic investigation into the mechanisms of resistance, are important for ensuring that current antibiotic treatment and prophylaxis recommendations remain relevant.

The fellow participated actively in all stages of the research study, starting from literature review and writing a study protocol to performing the laboratory analysis (culture of the meningococcal isolates, E-test, PCR assays for the *penA*, *rpoB* and *gyrA* genes, electrophoresis of the PCR products and purification, analysis of the electropherograms, use of the PubMLST.org/*Neisseria* database for assignment of alleles at antigenic loci), data entry, data analysis and correlation of phenotypic and genotyping susceptibility data. The results of this project were presented as poster at the 33rd European Congress of Clinical Microbiology and Infectious Diseases (ECCMID) 2023 (see 8.2.1) and orally at the 16th Congress of the European Meningococcal and Haemophilus Disease Society (EMGM) 2023 (see 8.2.3). The fellow was the first author of a manuscript published in 'Antibiotics' journal (see 8.1.1).

Educational outcome

The fellow participated in all stages of the research project, from planning a public health microbiology study, writing a project proposal and a study protocol, conducting the study, analysing data, using publicly accessible *Neisseria* database and interpreting as well as communicating the findings. This resulted in writing abstracts for international congresses, submitting them and presenting the results either orally or in a poster format. Also, the fellow familiarised herself with the whole process of preparing a manuscript for publication, from writing to addressing reviewers' comments successfully and publishing in a peer-reviewed journal.

3. Applied public health microbiology and laboratory investigations

A. Poliovirus Laboratory Surveillance at the Hellenic Pasteur Institute

Supervisor: Dr Emmanouil Angelakis.

Poliomyelitis (polio) is an endemic infectious disease caused by an RNA virus (poliovirus), belonging to the genus Enterovirus within the Picornaviridae family. It is a highly contagious virus that transmits via droplets or aerosols from the throat and by faecal contamination of hands, utensils, food and water, mostly affecting children under five years of age. Most poliovirus infections are asymptomatic, about 25% present mild symptoms and <1% of all infections present as paralytic poliomyelitis, affecting the brain (meningitis) or the spinal cord (acute flaccid paralysis - AFP). As humans are the only reservoir for poliovirus, global public health efforts to eradicate poliovirus are continuing through immunising every child until transmission of the virus has stopped, and the world becomes polio-free. AFP surveillance is the gold standard for detecting polio cases and essential for global polio eradication. All cases of AFP should be investigated for poliovirus as other enteroviruses (mainly types 70 and 71, echoviruses, and coxsackieviruses) can cause disease that mimics paralytic poliomyelitis. Greece was declared a 'polio-free area' in June 2002, along with the rest of Europe, as a result of systematic vaccination and enhanced epidemiological surveillance of polio. Surveillance of AFP in all persons under 15 years of age or in persons of any age when poliomyelitis is suspected, is a key element in maintaining a polio-free country and is performed at the National Poliovirus/Enterovirus Reference Laboratory (NPRL) - WHO accredited, in the Hellenic Pasteur Institute. AFP is investigated by molecular detection of enteroviruses

(Pan-Enterovirus Real-time RT-PCR) and polioviruses (Pan-Polio Real-time RT-PCR), isolation in cell culture (L20B, RD) and molecular typing (Real-time RT-PCR). Given that Greece continues to receive many short-term arrivals, students, migrants, and refugees from countries in which poliovirus is endemic, it is important to guarantee high-quality surveillance for maintaining its polio-free status until global eradication is achieved.

The fellow was exposed to the workflow of stool samples examination referred to NPRL for suspicion of AFP and based on this training activity she wrote a report (see 8.1.2.2) regarding the existing laboratory surveillance of poliovirus in Greece.

B. Genotypic testing of transmitted drug resistance in newly HIV-1 diagnosed treatment-naïve patients in Greece

Supervisors: Assoc. Prof. Apostolos Beloukas, Mrs Kassandra Prokter.

Baseline (before antiretroviral therapy – ART – initiation) HIV-1 RNA viral load quantification, and determination of resistance using nucleic acid-based molecular diagnostic assays are essential as standard of care for assessing prognosis and response to ART of people living with HIV (PLWH). The global scale-up of antiretroviral drugs has been accompanied by the emergence of HIV drug resistance (HIVDR), the levels of which have steadily increased in recent years. HIVDR can compromise the effectiveness of ARV drugs in reducing HIV incidence and HIV-associated morbidity and mortality and is emerging as a threat to the elimination of AIDS as a public health problem by 2030. All current antiretroviral drugs, including newer classes, are at risk of becoming partly or fully inactive due to the emergence of drug-resistant viral strains. HIVDR can be categorised as: a) acquired HIV drug resistance (ADR) when HIV mutations emerge because of viral replication among people receiving ARV drugs; and b) transmitted HIV drug resistance (TDR) when people are infected with HIV that harbours drug resistance mutations (DRM). Up to 10% of adults starting HIV treatment can have drug resistance to the non-nucleoside reverse transcriptase inhibitors (NNRTI) drug class. To minimise the emergence and transmission of drug resistant HIV, WHO recommends routine surveillance of HIVDR by genotypic resistance testing. The National HIV/AIDS Reference Centre for Southern Greece performs diagnosis and/or confirmation of HIV diagnosis. Furthermore, in newly HIV-1 diagnosed patients as well as in PLWH under ART treatment, quantification of HIV-1 viral RNA and routine genotypic HIV resistance testing are also performed. Routine surveillance of HIVDR in people initiating ART is of utmost importance, to inform optimal selection of first-line ART regimens, minimising treatment failure. Furthermore, surveillance of HIVDR provides evidence to adapt national treatment guidelines according to findings and optimise both patient and population-level treatment outcomes.

The fellow was exposed to the workflow of plasma blood samples referred to the National HIV/AIDS Reference Centre for Southern Greece for genotypic HIV resistance testing for protease (PR), reverse transcriptase (RT), and integrase in newly HIV-1 diagnosed patients. She also evaluated the received electropherograms herself using SeqScape™ software, MEGA software, and the Stanford University HIV Drug Resistance Database (HIVdb) and compared the results with the results obtained by the skilled personnel of the laboratory. Based on this training activity she wrote a report (see 8.1.2.5).

Educational outcome

The fellow applied concepts of virology to the public health disciplines and recognised the specific issues with the use of laboratory and epidemiological methods in investigations of rare and emerging diseases. She also identified the use and limitation of diagnostic and typing methods and their interpretation in patient diagnosis, surveillance, and epidemiological studies. She familiarised herself with the laboratory-based surveillance of both poliovirus and HIV drug resistance in her country, acquiring experience in molecular techniques used for enterovirus/poliovirus detection and assessment of HIV drug resistance and was exposed for the first time to cell line cultures for virus isolation.

4. Biorisk management

A. BSL-3 training at Hellenic Pasteur Institute

Supervisor: Dr Emmanouil Angelakis.

The fellow familiarised herself with biosafety and biosecurity practices in a Biosafety Level 3 (BSL-3) laboratory environment during the training activity at the BSL-3 laboratory of the Hellenic Pasteur Institute. Demonstration of the laboratory workflow and an introduction to biosafety, biosecurity practices and to laboratory best practice and technical procedures (GMPP) in a BSL-3 lab facility as well as discussion on specific laboratory characteristics was conducted. Also, specific topics were discussed; donning and doffing of Personal Protective Equipment (PPE), transfer and transportation of infectious substances, specimen receipt and storage, information on laboratory equipment with special emphasis on biosafety cabinets (BSCs), waste sterilisation and emergency/incidence report (contingency plan). The BSL-3 training was summarised in a report (see 8.1.2.3).

In addition to the BSL-3 training, the fellow conducted biosafety risk assessment in the context of the MDR-TB post-module homework assignment, after the Biorisk and Quality management module, in January 2022. The BIORAM Biosafety and Biosecurity software, provided by the Principal Expert Public Health Microbiology Training, was used.

Educational outcome

The fellow had the opportunity to read WHO Laboratory biosafety manual, fourth edition and refresh some skills pertinent to GMPP, transfer and transportation of infectious substances and decontamination. She also had a hands-on experience that included donning and doffing of PPE as well as using for the first time a Powered Air Purifying Respirator (PAPR). The fellow perceived the value of thorough knowledge of biosafety and biosecurity principles in handling highly pathogenic microorganisms in a BSL-3 laboratory setting as well as of hands-on training for ensuring proficiency of personnel in safe entrance into an operational BSL-3 laboratory. Furthermore, she understood the importance of biorisk management rules in controlling or minimising the risk to acceptable levels in relation to employees, the community, and others as well as the environment, which could be directly or indirectly exposed to biological agents or toxins.

5. Quality management

A. Quality management audit at the National Meningitis Reference Laboratory

Supervisors: Prof. Georgina Tzanakaki, Dr Athanasia Xirogianni.

Quality management audit at the National Meningitis Reference Laboratory was performed following a post-module homework assignment, after the Biorisk and Quality management module, in January 2022. The fellow had the opportunity to familiarise herself with a laboratory quality management evaluation using an evaluation tool provided by the Principal Expert Public Health Microbiology Training. The fellow conducted the audit in the National Meningitis Reference Laboratory, which is a laboratory accredited according to ISO:15189. The quality manager responded to all the questions regarding the process and quality control as well as documentation. Specifically, the questions addressing the process and quality control were about the accommodation and environmental conditions, the quality management and assurance, the pre-analytical process and specimen management, the analytical and post-analytical processes, and the improvement of quality. Furthermore, the questions about documentation concerned the following topics: quality document and document control, technical record, control of non-conformities, equipment logbooks, biosafety, personnel, stock, transport and surveillance/outbreak report documentation and standardised operating procedures/report format. This short assessment showed that the Documentation was excellent (96%) and the Process Management and Quality Control was very good (79%). The overall grade in the Process Management and Quality Control was higher considering that the laboratory performs Internal Quality Control regularly but not in each analysis. Overall, the NMRL is very well organised and satisfies the requirements of ISO 15189.

The fellow completed the audit assignment by interviewing laboratory personnel, going through protocols, methods, descriptions, and workflows in the laboratory.

B. Overall reassessment of the National Meningitis Reference Laboratory (NMRL) by the Hellenic Accreditation System

Supervisors: Prof. Georgina Tzanakaki, Dr Athanasia Xirogianni.

Accreditation of clinical and public health laboratories is the process by which an independent and authorised agency evaluates their quality system and competence based on certain pre-defined standards. ISO 15189 International Standard specifies requirements for competence and quality that are particular to medical laboratories. A medical laboratory's fulfilment of the requirements of this International Standard means that the laboratory meets both the technical competence requirements and the management system requirements that are necessary for it to consistently deliver technically valid results, improving the medical laboratory services which are essential to patient care. The Hellenic Accreditation System (ESYD) has been appointed by the Greek government as the National Accreditation Body of Greece according to the Regulation (EC) No 765/2008, assesses the technical competence, independence and impartiality of certification, inspection, and verification bodies as well as testing and calibration laboratories. The accredited status is valid for four years on condition that the Client meets the requirements on the interim inspections. An annual surveillance of the accredited organisation is carried out (on-site visit), while an overall reassessment of the laboratory is conducted every four years. The NMRL is accredited as a medical laboratory according to ELOT EN ISO 15189:2012. The overall reassessment of the NMRL took place on March 2022. The audit included questions and checks on both management and technical activities performed in the laboratory to ensure that all the procedures, from specimen arrival in the laboratory up to the communication of the results to who requested them, are included in a quality management system and adhere to it. Overall, the NMRL complied with the requirements of the ISO 15189. The audit confirmed the technical competence of laboratory personnel and their knowledge of the management system procedures relevant to their work. Only minor non-conformities were revealed by the audit. Possible corrective actions for implementation were discussed.

The fellow observed the overall reassessment of the NMRL and completed a report based on the laboratory audit (see 8.1.2.4).

Educational outcome

The fellow was familiar with audits and quality systems as she has been working in an ISO 17025 accredited public health laboratory. However, that was the first time she was exposed to a medical laboratory audit according to ISO 15189 and this experience helped her understand that the same principles apply to both ISO 17025 and ISO 15189, further consolidating her knowledge. The fellow found very interesting and educational the resolution of the non-conformities as well as the reasoning behind the proposed corrective actions.

6. Public health microbiology management

A. Tuberculosis laboratory network survey – 2021 update, with emphasis on molecular methods and COVID-19 pandemic impact

Supervisors: Dr Dimitrios Papaventsis, Dr Panayiotis Ioannidis, Dr Simona Karabela.

Before the COVID-19 pandemic, tuberculosis (TB) was the leading cause of death due to a single infectious agent. The COVID-19 pandemic has reversed years of global progress in tackling tuberculosis. Into this framework, the public health importance of the timely and accurate laboratory diagnosis of TB is considered a central component of any TB control program, as well as for the disease surveillance in Greece and in Europe. The aim of this project was to design, develop and implement National Survey of TB diagnostic services - 2021 update - in Greece through an on-line/electronic questionnaire using the most recent international standards and guidelines, to evaluate the capacity and differences in TB diagnostic services based on data from previous audits (2005 & 2016). A special emphasis was given on the use of molecular methods as the first diagnostic approach and on COVID-19 pandemic impact on TB laboratory services. Data returned were evaluated in terms of response rate, current laboratories capacity and active networking, implementation of recent WHO guidelines on rapid diagnostics for tuberculosis detection, impact of the COVID-19 pandemic in terms of laboratory performance and TB diagnostics and compliance with basic quality and biosafety guidelines. The collected data will be compared with previous relevant ones, to estimate the capacity trend over time. Furthermore, the survey was to function as a tool for the evaluation of the performance of the National Network for Tuberculosis, considering the impact of the recent COVID-19 pandemic. Finally, the results will be used in the context of the ongoing planning and implementation of a national strategic plan for the control of tuberculosis in Greece.

During this project, the fellow performed a literature review on current TB laboratory diagnostic standards, guidelines, protocols and algorithms, prepared the questionnaire, designed and implemented an online survey through EU Survey, performed data analysis, evaluation and interpretation. The results of this project were presented orally at the National Microbiology Conference 2022 (preliminary results) (see 8.3.1), as poster at the 33rd European Congress of Clinical Microbiology and Infectious Diseases (ECCMID) 2023 (see 8.2.2) and as oral presentations during the Project Review Module 2022 (see 8.3.2) and to the higher authorities of the NPHO administration 2022 (see 8.3.3).

B. Public Health microbiology management components as part of regular projects

Public Health microbiology management was an integral component of all projects and activities during the fellowship. This included laboratory management, ethical and integrity considerations, team working, research collaboration, time management, and working in several multidisciplinary teams with microbiologists, biologists, physicians, epidemiologists and laboratory technicians. The fellow's communication output in terms of manuscripts, reports and presentations is listed in section 8. The fellow interacted with different supervisors during her projects, while the changing of working environments among the Greek EUPHEM Consortium laboratories, gave the fellow insight into different leading strategies. Also, the presentation on 'Successful Public Health Leaders in time of crisis' at ECDC (see 8.3.5) was very educational and challenging.

Educational outcome

The fellow gained experience in working in a multidisciplinary public health team; exercised communication skills with different audiences, including higher authorities involved in decision-making for public health interventions, as well as public and media through simulation exercises. She understood how to construct a dedicated questionnaire and pilot testing it; acknowledged the need of team management and close collaborations in planning, scheduling and organising projects; realised the importance of laboratory database management ensuring security and integrity; and understood the role and responsibilities to be an inspiring leader and an effective manager within public health environment.

7. Teaching and pedagogy

A. Co-facilitation in a case study to MSc students at the School of Public Health, University of West Attica

Supervisor: Prof. Georgina Tzanakaki.

The case study 'Laboratory-confirmed invasive meningococcal disease in Saudi Arabia: effect of the Hajj vaccination policy, Saudi Arabia, 1995 to 2011' was used in a case study session for the Public Health MSc students during the lecture 'Outbreak management' in the frame of a series of lectures on 'Molecular epidemiology of nervous system, respiratory and sexually transmitted diseases' at the Department of Public Health Policy, School of Public Health, University of West Attica. The case study took place on 3 October 2021 virtually and lasted 2.5 hours. It was facilitated by Prof. Tzanakaki and the fellow co-facilitated. As the fellow was exposed for the first time to this type of training as a facilitator it was very educational for both the fellow and the students.

B. Lectures in the Postgraduate Program 'Public Health' of the School of Public Health, University of West Attica

The fellow is the Head of the Water Microbiology Laboratory at the Central Public Health Laboratory. As an expert on this field, the fellow is preparing and delivering lectures each year for the MSc students at the School of Public Health, University of West Attica, Greece.

After an invitation from the Assist. Prof. Georgia Mandilara, two lectures on 'Water Microbiology' took place virtually on 11 November 2021 and on 25 October 2022. They were given for the Public Health MSc students in the frame of a series of lectures on 'Surveillance of Food- and Waterborne Infections', Department of Public Health Policy, School of Public Health, University of West Attica.

C. Lecture in the Interdisciplinary Postgraduate Program 'Biotechnology' of the Department of Biology, National and Kapodistrian University of Athens

Supervisor: Prof. Panagoula Kollia.

A 2-hour lecture on 'Water Microbiology' given for the MSc students of the Interdisciplinary Postgraduate Program 'Biotechnology' of the Department of Biology, National and Kapodistrian University of Athens, in the context of the course 'Food Biotechnology and Nutrition' took place in person on the 26th of May 2022. The target audience consisted of 13 MSc students. The learning needs of the audience were assessed based on their occupational and educational needs and level of expertise, in collaboration with the Director of the Interdisciplinary Postgraduate Program, Prof. Kollia Panagoula. Teaching was conducted by Powerpoint presentation. At the end of the presentation, an anonymous questionnaire was distributed to the participating students to evaluate the lecture and based on their evaluation the fellow produced a reflective note (see 8.1.2.6).

D. Translation in Greek and facilitation of 2 case-studies to MSc (PH) students at the School of Public Health, University of West Attica

i) Supervisor: Assoc. Prof. Eleni Patsoula.

Facilitation of a case study was conducted regarding the principles of an outbreak investigation of an emerging vector-borne disease. The case study 'West Nile virus outbreak in Serbia, 2013' (adapted by Dr Mitra Drakulovich - National Institute of Public Health 'Dr Milan Jovanovic-Batut' Belgrade, Serbia - for MediPIET training purposes), was translated into Greek, adapted, and presented. 'The 2.5-hour teaching activity took place on-line in the frame of a series of lectures on 'Surveillance of Parasitic, Tropical, Zoonotic and Vector-borne Diseases', on the 11th of January 2023. The target audience consisted of 11 MSc students of the Postgraduate Program 'Public Health', specialisation 'Infectious Diseases – Public Health Laboratories'. The learning requirements of the audience were assessed based on their occupational and educational needs and level of expertise, in collaboration with Assoc. Prof. Patsoula. All participants actively participated by reading out loud one after the other the questions and background data and thereafter answering questions in turn. The students filled in an anonymous questionnaire and based on their evaluation the fellow produced a reflective note (8.1.2.7).

ii) Supervisor: Assist. Prof. Panagiota Giakkoupi.

Facilitation of a case study was conducted regarding the principles of the investigation of a healthcare associated infection (HAI) outbreak. The case study 'Outbreak of MRSA in a Neonatal Intensive Care Unit' (that was presented at Spetses in 2022 during the second part of the introductory course was translated into Greek, adapted, and presented. The two-hour teaching activity took place in person at the School of Public Health, University of West Attica on 21 January 2023, in the frame of a series of lectures on 'Molecular Epidemiology of Antibiotic Resistance and Nosocomial Infections'. The target audience consisted of 11 MSc students of the Postgraduate Program 'Public

Health', specialisation 'Infectious Diseases – Public Health Laboratories'. The learning requirements of the audience were assessed based on their occupational and educational needs and level of expertise, in collaboration with Assist. Prof. Giakkoupi. All participants actively participated by reading out loud one after the other the questions and background data and thereafter answering questions in turn. The students filled in an anonymous questionnaire and based on their evaluation the fellow produced a reflective note (8.1.2.8).

E. Translation in Greek and facilitation of a case-study to undergraduate students at the School of Health Sciences and Welfare, University of West Attica

Supervisor: Assoc. Prof. Apostolos Beloukas.

A three-hour teaching activity was conducted in person at the University of West Attica on 6 April 2023, in the frame of a series of lectures on 'Food and water microbiology'. The target audience consisted of seven undergraduate students of the Laboratory of Molecular Microbiology and Immunology, Department of Biomedical Sciences, School of Health Sciences and Welfare. After assessment of the learning requirements of the audience in collaboration with Assoc. Prof. Apostolos Beloukas a teaching activity consisting of two lectures on 'Waterborne infectious diseases – outbreaks' and '10 steps of an outbreak investigation' followed by facilitation of the case study: 'Giardiasis in Bergen, Norway, September – December 2004' was performed by PowerPoint. The case study was translated into Greek, adapted, and presented so the audience could 'learn by doing'. At the end of the activity, the students filled in an anonymous questionnaire and based on their evaluation the fellow produced a reflective note (see 8.1.2.9).

Educational outcome

The fellow learned and gained experience in defining learning objectives based on the needs of different multidisciplinary audiences, designing, and preparing various learning materials (interactive lectures and case studies) and delivering and evaluating the learning activities.

8. Communication

8.1 Publications related to the EUPHEM fellowship

8.1.1 Manuscripts published in peer-reviewed journals

1. Spiliopoulou I, Xirogianni A, Simantirakis S, Tzanakaki G. Meningococcal Antibiotic Resistance: Molecular Characterization of Isolates from Patients with Invasive Meningococcal Disease (IMD) in Greece. *Antibiotics*. 2023;12(7):1136. doi:10.3390/antibiotics12071136. <https://www.mdpi.com/2079-6382/12/7/1136>

Publications in preparation

1. Ioanna Spiliopoulou, Danai Pervanidou, Anastasia Mpimpa, Nikolaos Tegos, Eleni Patsoula. Evaluation and further analysis of genotyping *P. vivax* samples from malaria cases recorded in Greece during 2015-2019. (in final preparation)

8.1.2 Other reports

1. Report on a community waterborne outbreak due to *Salmonella* Bovismorbificans, Greece, August 2022.
2. Training activity report on Poliovirus laboratory surveillance at the Hellenic Pasteur Institute.
3. BSL-3 training activity report at the Hellenic Pasteur Institute.
4. Laboratory audit report of the National Meningitis Reference Laboratory.
5. Training activity report on laboratory surveillance of pretreatment Human Immunodeficiency Virus (HIV) Drug Resistance.
6. Reflective note on teaching_1
7. Reflective note on teaching_2
8. Reflective note on teaching_3
9. Reflective note on teaching_4

8.2 Conference presentations

1. Ioanna Spiliopoulou, Athanasia Xirogianni, Stelmos Simantirakis, Georgina Tzanakaki. Antimicrobial resistance of *Neisseria meningitidis* strains isolated from patients in Greece, 2010–2021. 33rd European Congress of Clinical Microbiology and Infectious Diseases (ECCMID), 15–18 April 2023, Copenhagen, Denmark, poster presentation (P1786).
2. Ioanna Spiliopoulou, Dimitrios Papaventsis, Panayotis Ioannidis, Simona Karabela. Impact of the COVID-19 pandemic on tuberculosis public healthcare laboratory diagnostic services, Greece, 2019–2021. 33rd European Congress of Clinical Microbiology and Infectious Diseases (ECCMID), 15–18 April 2023, Copenhagen, Denmark, poster presentation (P1107).
3. Ioanna Spiliopoulou, Athanasia Xirogianni, Stelmos Simantirakis, Georgina Tzanakaki. Molecular characterization of antibiotic susceptibility of *Neisseria meningitidis* isolates from patients with invasive meningococcal disease in Greece from 2010 to 2021. 16th Congress of the European Meningococcal and Haemophilus Disease Society (EMGM), 29 May to 1 June 2023, Dubrovnik, Croatia, oral presentation.
4. L. Politi, T. Sideroglou, A. Chrysostomou, G. Mandilara, I. Spiliopoulou, A. Theofilou, A. Bordhoka, S. Poufta, K. Mellou, T. Zaoutis, D. Paraskevis. A community waterborne outbreak due to *Salmonella* Bovismorbificans, Greece, August 2022. European Scientific Conference on Applied Infectious Disease Epidemiology (ESCAIDE), 22–24 November 2023, Barcelona, Spain, poster presentation.

Abstracts submitted (not accepted)

- a) I. Spiliopoulou, D. Papaventsis, P. Ioannidis, S. Karabela. Impact of the COVID-19 pandemic on tuberculosis public healthcare laboratory diagnostic services, Greece, 2019–2021. European Scientific Conference on Applied Infectious Disease Epidemiology (ESCAIDE) 2022.
- b) I. Spiliopoulou, D. Papaventsis, P. Ioannidis, S. Karabela. Implementation of WHO-recommended rapid diagnostic (mWRD) tests for tuberculosis in Greece: 2021 national survey data. European Scientific Conference on Applied Infectious Disease Epidemiology (ESCAIDE) 2022.

8.3 Other presentations

1. Spiliopoulou Ioanna. Network of tuberculosis diagnostic laboratories in Greece - Latest data in the era of the COVID-19 pandemic. National Microbiology Conference, 28–30 January 2022, Athens, Greece, oral presentation.
2. I. Spiliopoulou, D. Papaventsis, P. Ioannidis, S. Karabela. Impact of the COVID-19 pandemic on tuberculosis public healthcare laboratory diagnostic services, Greece, 2019–2021. Oral presentation during the Project Review Module. 26 August 2022, Lisbon, Portugal.
3. Spiliopoulou Ioanna. Network of tuberculosis diagnostic laboratories in Greece - Latest data in the era of the COVID-19 pandemic. Oral presentation of the results of the national TB survey to the NPHO administration. 29 September 2022, NPHO, Athens, Greece.
4. Duro D, Papageorgiou G, Karaj G, Tragari D, Spiliopoulou I, Sideroglou T, Chrysostomou A, Polemis M, Mellou k, Mandilara G. Laboratory investigation of a waterborne *Salmonella enterica enterica* ser. Bovismorbificans outbreak. 13th Panhellenic Conference of the Panhellenic Association of Bioscientists (PEV), 9–12 December 2022, Thessaloniki, Greece, oral presentation.
5. Spiliopoulou Ioanna and Corrado Minetti. Successful Public Health Leaders in time of crisis. Oral presentation of Group's 4 work to ECDC Director, Chief Microbiologist and Chief Scientist during the MLCPH module. 12 May 2022, ECDC, Stockholm.
6. I. Spiliopoulou. Evaluation and further analysis of genotyping *P. vivax* samples from malaria cases recorded in Greece during 2015–2019. Oral presentation during the Project Review Module. 28 August 2023, Lisbon, Portugal.

9. EPIET/EUPHEM modules attended

1. Fellowship Introductory Course Part 1, 20 September to 8 October 2021, virtual
2. Fellowship Introductory Course part 1, Inject Day 1 Phylogeny & WGS, 20 October 2021, virtual
3. Fellowship Introductory Course part 1, Inject Day 2 Operational Research, 27–28 October 2021, virtual
4. Fellowship Introductory Course part 1, Inject Day 3 Data Collection and Management, 10–11 November 2021, virtual
5. Outbreak Investigation Module (OIM), 6–10 December 2021, virtual
6. Biorisk and Quality Management module, 17–18 January 2022, virtual
7. Multivariable Analysis (MVA) module, 14–18 March 2022, virtual
8. Multivariable Analysis (MVA) module, Inject Day, 30 March 2022, virtual
9. Fellowship Introductory Course 2021 part 2, Project Review Activity, 20–22 April 2022, Spetses, Greece
10. Fellowship Introductory Course 2021 part 2, 25–29 April 2022, Spetses, Greece
11. Rapid Assessment and Survey Methods (RAS) module, 6–10 June 2022, virtual
12. Project Review Module 2022, 29 August to 2 September 2022, Lisbon, Portugal
13. Time series analysis (TSA) module, 7–12 November 2022, Bilthoven, the Netherlands
14. Qualitative Research (QR), Optional Inject Days, 31 January and 3 February 2023, virtual
15. Vaccinology module, 13–17 February 2023, virtual
16. Management, Leadership and Communication in Public Health (MLCPH) Module, 8–12 May 2023, Stockholm, Sweden
17. Project Review Module 2023, 28 August to 1 September 2023, Lisbon, Portugal

10. Other training

1. Online course of the United Nations Department of Safety and Security (UNDSS), May 2022. BSAFE certificate of achievement
2. Webinar organised by IDEXX on 'Protecting public health from the threat of Legionnaires' disease', 8 September 2022
3. Webinar organised by ESCMID on 'Genomic surveillance of viral pathogens: are we prepared for emergent global threats across One Health', 20 September 2022
4. Webinar organised by ESCMID on 'Comparison of new diagnostic techniques multiplex PCR and WGS enteric infection surveillance with conventional diagnostic procedures', 4 October 2022
5. Webinar organised by ESGEM on 'Using genomics for antimicrobial resistance at the public health and clinical interface', 11 October 2022
6. Webinar organised by Epiet Alumni Network (EAN) on 'Xenomonitoring and surveillance: Using mosquitoes to find (and control) pathogens', 9 January 2023
7. Webinar organised by Novogene on 'A Beginner's Guide to Next Generation Sequencing: Principles & Recent Developments', 19 January 2023
8. Webinar organised by Labroots 'Real-time Whole Genome Sequencing Surveillance for Healthcare Outbreak Detection and Investigation', 28 February 2023
9. Webinar organised by 'Ippokrateio' General Hospital of Athens on 'Management of biological hazards and laboratory biosecurity', 1 March 2023
10. Webinar organised by EAN entitled 'Mobile laboratories in public health interventions', 4 May 2023

11. Other activities

1. Spiliopoulou I, Aivalioti K, Theofilou A, Tryfinopoulou K, Vatopoulos A. Microbiological quality of dialysis water and dialysis fluid samples in a 7-year period: the Central Public Health Laboratory experience. Panhellenic Conference on Public Health 2022, 28 February to 2 March 2022, Athens, Greece, oral presentation.
2. Spiliopoulou I. Network of Public Health Laboratories of the National Public Health Organization (Central and Peripheral Public Health Laboratories). Meeting 'SAFE WATER' organised by the Environmental Microbiology Unit of the Department of Public Health Policy, School of Public Health, University of West Attica and the Panhellenic Association of Public Health Supervisors, 20 May 2022, Athens, Greece, oral presentation.
3. Spiliopoulou I. Requirements for the microbiological testing of water intended for human consumption and interpretation of the microbiological analysis results. Meeting 'SAFE WATER' organised by the Environmental Microbiology Unit of the Department of Public Health Policy, School of Public Health, University of West Attica and the Panhellenic Association of Public Health Supervisors, 20 May 2022, Athens, Greece, oral presentation.
4. Participating in an on-line meeting with the General Director of the Ministry of Health concerning the transposition of the new EU Directive 2020/2184 on the quality of water intended for human consumption in the national legislation with special concern on the microbiological parameter of *Legionella*, 27 October 2022.

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