



D6.14: Report on Short Term Missions 2021 (Y4)

Work Package 6

Responsible Partner: UoS, UK (P23)



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REPORT ON SHORT TERM MISSIONS Y4

Introduction

Short Term Missions (STMs) are small co-funded travel grants which offer and exchange scientific expertise, methodologies, equipment, and facilities to our consortium members. The aim of these missions is to harmonise the existing approaches and methodologies within the One Health EJP (OHEJP).

These STMs drive research forward in a collaborative and non-duplicative fashion to strengthen the scientific capacity within the OHEJP, and to also contribute to the future prevention, preparedness, detection, and response of the EU to foodborne and other emerging threats across human-animal-environmental sectors.

The OHEJP is able to co-fund up to ten STMs each year (each funded 44% by the EU). The Education and Training activities team from Work Package 6 (WP6) are responsible for co-ordinating the call, selection of applicants for funding (through a review process) and reporting on the short term missions funded each year.

Short Term Missions 2021 call

The call was launched on 13th January 2020, and a promotional email marketing strategy was used to disseminate the call information to the Scientific Steering Board (SSB), Project Management Team (PMT), Programme Managers Committee (PMC), Project Leaders, PhD teams, and Communication Contact Persons. The email was visually attractive and incorporated a branded launch infographic and branded text.

The email contained instructions on how to access the 'Call for Short Term Missions 2021' group on the private space of the OHEJP website, where they could download the guidelines, application form and templates. Readers were asked to forward the email to those in their institute who may find it useful.

The launch of the call was also disseminated on the Education and Training Open Calls page of the OHEJP website in the form of call flyers containing interactive links to the guidelines and application forms. This page was promoted in the Education and Training activities monthly bulletins, the OHEJP consortium newsletter, and social media channels, Twitter and LinkedIn, to increase traffic to the website.

Selection of Short Term Missions

The WP6 Team co-ordinate the call and selection of the STMs following the validated procedure which involves the following steps:

Applicants must fill in the STM application form, and are strongly encouraged to compile the following documents with the main application: a detailed work plan, financial budget template, letters of recommendation from the home and hosting institute, *Curriculum Vitae*, and finally a list of publications (if applicable). After the deadline closed, the WP6 Team performed an eligibility check with pre-determined eligibility criteria.



Eligibility criteria included that only scientific staff from, and PhD students based at, the partner institutes of the OHEJP were eligible to apply. STM applications would only be considered if they relate to the key OHEJP priority areas: One Health Missions- Veterinary, Food, Medical and or Environmental research; skills development missions (e.g. genomics, bioinformatics, big data epidemiology); exchange with researchers, policy makers, and risk managers to complement WP5- Science to Policy translation; risk research; integration of microbiological, risk assessment and surveillance activities; and finally, harmonisation of diagnostics tests, platforms and research. There was no limit for the duration of the STM, as long as the funds requested adhere to the OHEJP budgetary rules, and STMs could take place between Jan and Dec 2021.

Each application and supporting documents were sent to three independent reviewers. The reviewers were nominated by the SSB.

The WP6 Team compiled and reviewed the scores, detecting and resolving any typographical or administrative errors before submitting to the PMT. The 4 applications submitted were all eligible and of high quality. WP6 suggested a decision to PMT based on the actual comments of reviewers and list of missions recommended for funding. This decision to fund the 4 missions was approved by the PMT.

The STMs were awarded on the 21st September 2020, and the final decision was communicated to the SSB and applicants, and published on the OHEP website on a dedicated webpage for Short Term Missions 2021- <https://onehealthjep.eu/short-term-missions-2021/>.

Impact of COVID-19

The COVID-19 pandemic affected the success of these missions due to the disruption to travel plans and local restrictions. Two of four STMs took place in 2021. Of the remaining two STMs, one mission was cancelled (staff left the host institution) and the remaining one has been postponed (and scheduled) in 2022.

Short Term Missions

2. Short Term Mission 1

2.1. Short Term Mission 1 Report

Start-up of an efficient sequencing facility

Name of applicant

Cathrine Arnason Bøe

Institute of Affiliation/

Institute: The Norwegian Veterinary Institute



Contact information

Address: Elizabeth Stephansens vei 1, 1433 Ås, Norway

Email: cathrine.arnason.boe@vetinst.no

Host institute and names of scientists involved in STM

Statens Serum Institutt
Mette Theilgård Christiansen (initial contact)
Eva Møller-Nielsen
Christina Wiid Svarrer

Dates of STM

15th to 19th of November 2021

Call Topic

Short term missions 2021

Research Domain

Foodborne Zoonoses, AMR and Emerging threats

Key aims of STM or Workshop

The key aims of the STM were:

- to develop competence regarding how to efficiently run a sequencing facility/workflow through the week/operation of sequencers and robots
- exchange experiences regarding extraction of nucleic acids
- exchange experiences regarding sequencing of viral genomes.

Impact and relevance of scientific mission

Competence development concerning management of a sequencing lab and implementation of novel equipment to achieve a more automated workflow.

Benefits to OHEJP

The STM strengthened the collaboration between partners in a One Health aspect to address existing and emerging threats of antimicrobial resistance and zoonotic agents.

It also helped to harmonise the high throughput sequencing method between institutes in the One Health EJP project by training of Cathrine Arnason Bøe in an NGS facility with experience in routine sequencing for diagnostic, outbreak investigations and research purposes.

Summary

The STM to visit a sequencing facility at SSI, Copenhagen, Denmark was planned to take place during early winter 2021 but was postponed, due to the pandemic, until mid-November 2021. During the stay, routine WGS sequencing of bacteria and virus (SARS-CoV-2) was performed in the lab and in addition, several one on one meetings with key personnel for sequencing were held. The aims of the STM were to develop competence on how to efficiently run a sequencing facility, including how to optimise workflow through the week and to increase knowledge on operation of sequencers and robots. In



addition, exchange of experience regarding extraction of nucleic acids and sequencing of viral genomes was desired. The Norwegian Veterinary Institute (NVI) has an equipment platform set-up similar to SSI, but has not fully exploited its potential yet as the equipment for WGS was acquired recently. During the STM ideas and inspiration of how to improve routine sequencing and start novel sequencing tasks was acquired. Improvements regarding how to make use of the pipetting robots for more steps, simplify the quality control of sequence data and decision making on Nanopore vs Illumina sequencing for high risk agents like the avian flu are more specific examples of significant knowledge brought back to the sequencing facility at NVI. The STM was a revelation for all the possibilities we now have at NVI to implement more high throughput sequencing for surveillance, preparedness, diagnostics and research in the near future.

Technical Report

Background:

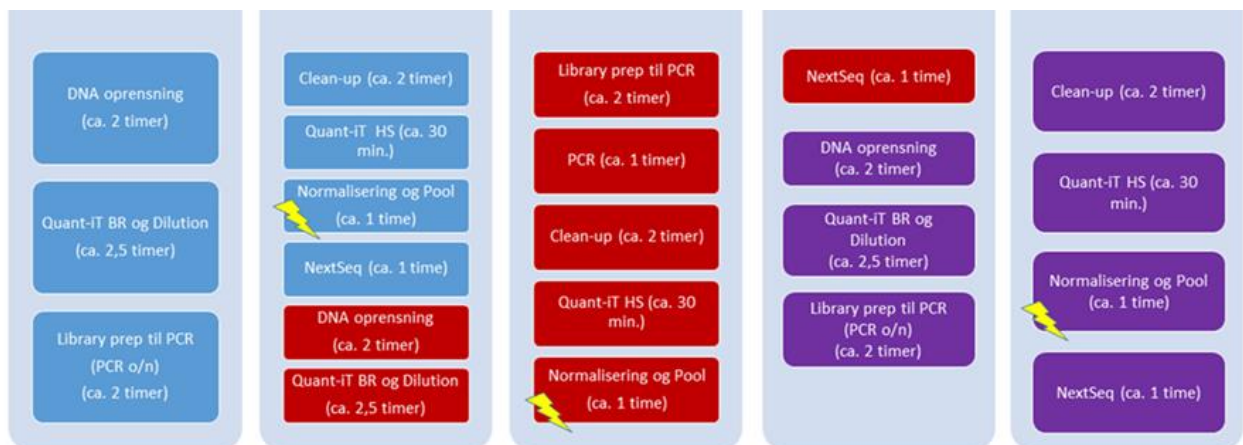
The project SEQ-TECH at The Norwegian Veterinary Institute (NVI) has acquired a platform of sequencing machines, pipetting robots and other equipment through 2019-2021 and is now set-up for an automated flow-through of several hundred high throughput sequencing samples per week. Although the equipment is there, the routine workflow is not fully established and the potential of the different machines has yet to be exploited. The STM applicant aimed to visit SSI and follow their weekly routine handling library preparation and high-throughput sequencing (HTS) of up to 300 samples per week. Their sequencing service resembles the platform SEQ-TECH is establishing.

Typical workflow for one week at SSI look like this:

Monday



Friday



Task 1:

During the STM one round of bacterial WGS from DNA extraction through sequencing on the NextSeq was followed. SSI are heavily involved in WGS of corona virus so one round of close to 800 SARS-CoV-2 samples undergoing library preparation and sequencing (NextSeq) was also covered. After DNA sequencing the data goes through a quality control by bioinformaticians. Extensive walkthrough of the quality control process was performed.



Task 2:

Meetings with Jesper Schak Krog, involved in sequencing of influenza virus (both Illumina and Nanopore platforms), and Søren Overballe-Petersen, involved in Nanopore sequencing and the project FULL-FORCE (EPJ One Health), was held to exchange experiences regarding areas of sequencing that SEQ-TECH is involved in or about to embark on.

Findings and conclusion:

Inspiration and several ideas of how to improve the routine flow, Hamilton pipetting robot usage and bioinformatics quality control end point at NVI was gained by observing the library prep and sequencing hands on in the lab and also the quality control pipeline at SSI during the STM. In addition, sharing of experiences regarding Nanopore sequencing and WGS of influenza virus during the two meetings (Task 2) will be valuable going forward.

List of dissemination and communication activities

Not applicable

Scientific outputs

Not applicable

Testimonial

This short-term mission allowed me to follow the routine whole genome sequencing of both bacteria and virus at the sequencing facility at Statens Serum Institutt (SSI). All steps starting from DNA extraction through sequencing and quality control of the resulting output data were covered. The experience was very useful and highly motivating before continuing implementing a more automatic workflow of high-throughput sequencing at The Norwegian Veterinary Institute. I will definitely use the new knowledge to advance and streamline several steps of our ongoing sequencing projects and routine workflows. In addition, the STM facilitated one to one meetings with researchers involved in collaborating projects. These meetings were especially beneficial for our planned surveillance projects of some zoonotic agents in addition to our work for preparedness and emerging threats. The connections made with researchers and technical staff during the STM will be valuable for the future. Returning from the STM I am very grateful to the OHEJP for granting this opportunity. I would also like to express gratitude to SSI and the kind staff for allowing me into their labs and showing me hands-on their efficient sequencing facility even though they had their hands full of SARS-CoV2 whole genome sequencing at that moment.



2.2. Short Term Mission 1 Case study

SHORT TERM MISSIONS

Short Term Missions (STMs) are small travel grants with the aim of:

- Sharing scientific expertise, methodologies, equipment and facilities to harmonise the existing approaches and methodologies within the large OHEJP European network
- Driving the research forward in a collaborative and non-duplicative fashion to strengthen both the scientific capacity within the OHEJP
- Contributing to the future prevention, preparedness, detection and response of the EU to foodborne and other emerging threats across human-animal-environmental sectors.

Image: Peter Artymiuk

Start-up of an efficient sequencing facility



Theme: Foodborne Zoonoses, AMR and Emerging threats
Home Institute: [Norwegian Veterinary Institute](#), Norway
Mission Hosting Institute: SSI, Denmark
Duration of Mission: 1 week



I will definitely use the new knowledge to advance and streamline several steps of our ongoing sequencing projects and routine workflows. The connections made with researchers and technical staff will be valuable for the future. I am very grateful to the OHEJP for granting this opportunity. I would also like to express gratitude to SSI and the kind staff for allowing me into their labs."

Cathrine Arnason Bøe,
Norwegian Veterinary Institute

The aim of this mission was to develop skills on the management of a sequencing facility using an automated workflow. The project SEQ-TECH at the Norwegian Veterinary Institute (NVI) has recently acquired equipment to set-up an automated high throughput sequencing platform. However, the routine workflow is not yet entirely established, and the potential of the different machines not fully exploited. By visiting the SSI and following their weekly routine for library preparation and high-throughput sequencing (HTS), the participant aimed to learn how to improve the NVI workflow.

During this mission, routine whole genome sequencing (WGS) of bacteria and virus (SARS-CoV-2) were performed, the data quality control process was described and one-on-one meetings with key personnel were held. Inspiration and several ideas of how to improve routine sequencing and start novel sequencing tasks was gained by observing the SSI workflow and quality control pipeline. In addition, sharing of experiences regarding Nanopore sequencing and WGS will be valuable going forward.

The STM was a revelation for all the possibilities available at NVI, using the new equipment, to implement higher throughput sequencing for surveillance, preparedness, diagnostics and research in the near future.

One Health EJP has received funding from the European Union's Horizon 2020 research and innovation programme under grant agreement No 773830.

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Figure 1: This case study can be found on the OHEJP website: <https://onehealth.ejp.eu/short-term-missions-2021/>



2. Short Term Mission 2

CarbaPlasmid – Tracking endemic carbapenemase plasmids in human, animal and environmental isolates

2.1 Short Term Mission 2 Report

Name of applicant	Liam Burke
Institute of Affiliation/	Institute: National University of Ireland Galway
Contact information	Address: Orbsen Building National University of Ireland Galway Galway, Ireland. Email: liam.burke@nuigalway.ie
Host institute and names of scientists involved in STM	VISAVET-Universidad Complutense Madrid Prof Bruno Gonzalez-Zorn
Dates of STM	4th – 30th July 2021
Call Topic	One Health Missions, Skills Development Missions
Research Domain	Antimicrobial resistance
Key aims of STM or Workshop	Skills Aim: <ul style="list-style-type: none"> Develop skills in hybrid sequence analysis for plasmid characterisation. Research aim: <ul style="list-style-type: none"> Understand the molecular epidemiology of carbapenemase plasmids circulating in Enterobacterales isolates in Ireland from a One Health perspective.
Impact and relevance of scientific mission	This mission developed capacity for long-read sequencing and hybrid sequence assembly in Irish OHEJP partner (NUI Galway), which will also be used to support the surveillance function of the Irish National CPE Reference Laboratory. This expertise will be fundamental in ongoing and future collaborative AMR research projects with OHEJP partners and



Benefits to OHEJP

in the surveillance of endemic carbapenemase plasmids in a One-Health context in Ireland.

The mission contributes to harmonisation of the approaches and methodologies for molecular epidemiological investigation of AMR platforms in a One Health context across One Health EJP partners, thereby strengthening the scientific capacity within the OHEJP. It strengthened collaborations between OHEJP partners NUI Galway and Visavet-UCM. It strengthens future prevention, preparedness, detection and response of the EU to emerging AMR threats across human-animal-environmental sectors.

Summary

Carbapenemases are enzymes produced by bacteria that inactivate carbapenems, antimicrobials of last resort which are used to treat multi-drug resistant infections. Carbapenemase Producing Enterobacterales (CPE) are a problem in hospitals and long-term care facilities, where they can cause outbreaks in inpatient populations. Such outbreaks are difficult to recognise and contain, as the carbapenemase genes that confer resistance are often located on mobile resistance plasmids, small circular pieces of DNA that bacteria share between each other in order to survive in areas where antimicrobials are present. Identifying plasmid spread between bacteria that cause these healthcare associated infections depends on the ability of medical scientists to recognise the sequence of the plasmids in different strains and species. This is extremely difficult, even with whole genome sequencing, as plasmids contain many repeat units that are difficult to resolve in the typical short-read assemblies used by reference labs. Resolving these repeats can be accomplished using longer, third-generation sequencing reads, such as those obtained using the Oxford Nanopore MinION sequencer, which are long enough to span most repeated regions. Hybrid assembly (assembly of long and short reads simultaneously) enables the accurate reconstruction of complete resistance plasmid sequences. This strategy has great potential for the investigation of resistance plasmid epidemiology, including from a One Health perspective.

The aim of this mission was to develop skills in Nanopore sequencing and hybrid sequence analysis in order to characterise antimicrobial resistance plasmids for One Health epidemiological investigations. In this project CPE isolated from the natural environment, hospital wastewater, the hospital environment and hospital patients in Galway, Ireland, were analysed by hybrid sequencing analysis. Several techniques and applications for analysis of hybrid bacterial sequence data were learned. A harmonised hybrid sequence analysis pipeline was successfully transferred between One Health EJP partners UCM and NUI Galway. A total of 39 bacterial genomes were fully resolved, including plasmids and chromosomes bearing the carbapenemase genes NDM-5, KPC-2 and OXA-48. This mission developed capacity for long-read sequencing and hybrid sequence assembly in Irish OHEJP partner NUI Galway, which will also be used to support the surveillance function of the Irish National CPE Reference Laboratory. This expertise will be fundamental in ongoing and future collaborative AMR research projects with



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OHEJP partners and in the surveillance of endemic carbapenemase plasmids in a One-Health context in Ireland.



Technical Report

Background

WGS is usually done via generation of high throughput next-generation-sequencing (NGS) reads, (e.g. Illumina MiSeq) which are typically short (<300 bp) but accurate (>98%). However mobile resistance genes residing within plasmids are often carried on transposable elements, repeated sequences that can be thousands of base pairs in length, which can cause problems during assembly. Resolving these repeats can be accomplished using longer, third-generation sequencing reads, such as those obtained using the Oxford Nanopore MinION sequencer, which are long enough to span most repeated regions. Recent advances in the reproducibility and accuracy of long-read sequencing, combined with reduced costs, have enabled more widespread use of this technology. Hybrid assembly (assembly of long and short reads simultaneously), although still not economically viable for routine use in the reference laboratory, enables the accurate reconstruction of complete resistance plasmid sequences. This strategy has great potential for the investigation of resistance plasmid epidemiology, including from a One Health perspective. Furthermore, once resolved by hybrid assembly, plasmid MLST schema can be designed to identify locally endemic resistance plasmids using NGS reads alone.

NGS data is available for environmental CPE isolates collected by our research group since 2016 from Irish sewage, wastewaters and natural waters including rivers, lakes and coastal seas. Environmental isolates producing NDM-5 (seawaters, lakes), OXA-48 (seawaters, estuary, rivers, sewage, wastewaters) and KPC-2 (seawaters, sewage) have been identified. NGS data is also available for every CPE isolate collected in Irish hospitals since 2018 and many prior to this. This data demonstrates frequent detection of the above CPE markers in clinical samples and screening samples from inpatient faecal swabs and hospital environments.

Aim:

This mission aims to characterise and compare similar carbapenemase plasmids of interest from these sources in order to infer possible dissemination routes, investigate plasmid evolution over time and to create a spatiotemporal One Health map of important carbapenemase plasmids.

Outcome:

The data will contribute to bridging the knowledge gaps that exist in our understanding of the role of the environment in the persistence, evolution and transmission of AMR plasmids and the propensity for carbapenemase plasmid transfer between different bacterial species and strains in distinct ecological niches. It may also identify emerging AMR threats from a One Health perspective.

Work plan

Preparation for mission:

Training in genomics data science analysis methods was undertaken from January to June 2021. Dr Burke completed a training course in bioinformatics locally at NUI Galway, and



gained a basic understanding of short and long read sequence assembly pipeline bioinformatics tools and AMR databases and became familiar with using the command line in Ubuntu. A collection of Carbapenemase Producing Enterobacterales isolates (including *Klebsiella* spp., *Escherichia coli*, *Citrobacter* and *Enterobacter* spp.) collected from natural waters (rivers, lakes and seas) were curated. Environmental CPE isolates producing KPC-2, NDM-5 and OXA-48 were chosen. Dr Niall Delappe, Medical Scientist in the National CPE Reference Laboratory (NCPERL) at University Hospital Galway collaborated to identify CPE isolates of interest with matching carbapenemase genes from the hospital environment, hospital clinical samples and inpatient screening samples. All 41 CPE isolates were collected between July 2018 and December 2020 in Galway City. With the assistance of the NCPERL, the isolate collection was sub-cultured to agar stabs and shipped to Prof Gonzalez-Zorn's Antimicrobial Resistance Unit (ARU) lab in UC Madrid. Illumina (short) sequence reads (previously sequenced), phenotypic data and metadata on the collection were curated in a cloud server database.

Week 1 and 2:

The first 9 isolates (containing KPC-2 and NDM-5 carbapenemase genes) were cultured and pure genomic DNA was isolated for sequencing using a Qiagen DNA prep kit. DNA concentration was normalised via Qubit fluorometric quantitation (Invitrogen). Sequencing libraries were prepared using the Ligation Barcoding Sequencing kit (Oxford Nanopore). The concentration for each barcode was checked via Qubit and libraries were pooled in equimolar concentration. Sequencing was run for approximately 48 hours on a flow-MIN106 R9 flow cell using a Nanopore MinION device. Basecalling was performed simultaneously using MinKnow resulting in raw FastQ sequencing data. Data files for each isolate were concatenated and adapters removed using Porechop. Quality control was done using Filtlong, removing all reads shorter than 1000 bp and the worst 5% of reads. Read depth was calculated for each isolate using a custom Python script to inform choice of assembly pipeline and ranged from 8 to 130x coverage. As most isolates had shallow Nanopore read depth (0-50x coverage), but deep Illumina read depth, Unicycler was chosen as the pipeline for hybrid assembly. The input was both Illumina reads (following quality control using fastp) and Nanopore reads. Assembly polishing used Pilon. Hybrid assemblies were visualised using Bandage. Assemblies were searched using BLAST against the resfinder database for antimicrobial resistance genes to identify the genomic location of the carbapenemase and other resistance genes of interest. Carbapenemase containing plasmid and genomic sequences were saved as separate FASTA files for further annotation and analysis. Plasmid finder and resfinder were used to identify plasmid replicons and antimicrobial resistance genes. Copla was used to identify the plasmid taxonomic unit (PTU) of carbapenemase plasmids. Bakta was used to annotate the sequences, which were transferred to Geneious Prime platform for further annotation and analysis.

Week 2 and 3:

The above methodology was repeated for the next run of 12 isolates (containing NDM-5 and OXA-48 carbapenemase genes).

Week 4



The Native Barcoding Expansion kit was used to sequence the last 20 isolates (all OXA-48), and one KPC-containing isolate that was not fully resolved during the first run was repeated. Bioinformatics analysis was carried out as described above.

Findings:

From the first sequencing run 8 of 9 assemblies were successful, resulting in complete genomes and circularised plasmids, as visualised in Bandage. Run two produced ten perfectly resolved genomes. The third sequencing run was not as successful - low DNA yields during library preparation meant only 15 of the 21 libraries were at sufficient concentration for sequencing. This was not unexpected, as it was the ARU laboratory's first time using the Native Barcoding Expansion kit, which potentially allows the sequencing of 24 isolates on a single flow cell. Sequencing failed for one of the 15 isolates, but complete hybrid assemblies were generated for the other 14.

The bioinformatics pipeline used was very successful in generating complete hybrid assemblies with shallow coverage long read sequences when combined with deep coverage short reads. Several new bioinformatics tools were also used for the first time by Prof Gonzalez-Zorn's group during this project, including those for bacterial sequence annotation (Bakta) and plasmid analysis (Copla).

A final sequencing run was planned for September in NUI Galway, but was delayed due to a University-wide cyberattack which made it impossible to connect to the wired internet and several IT and computing infrastructures on campus for several months. The final sequencing run was completed in late December 2021, resulting in complete hybrid assemblies for 7 of the 9 repeated isolates, and 2 partial assemblies which included fully circularised carbapenemase plasmids. This work demonstrated the success of the training and methods harmonisation elements of this STM, as the pipeline and sequencing protocol was transferred to NUI Galway and was validated by completion of hybrid sequencing of the isolates that failed initially in UCM. Altogether 39 complete hybrid genome assemblies were obtained, with chromosomally encoded and mobile resistance elements characterised for all isolates.

We resolved 5 KPC-2 containing carbapenemase plasmids of PTU FK, ranging in size from 115 to 317 kb. Two environmental *Klebsiella pneumonia* ST88 isolates harboured KPC-2 chromosomally. All 12 NDM-5 producing isolates harboured plasmid-encoded carbapenemase resistance, with plasmids ranging in size from a 46 kb PTU-X3 plasmid to a 148 kb PTU-FE plasmid. OXA-48 was also usually plasmid encoded, and associated with PTU-L/M plasmids ranging in size from 48 to 85 Kb. However one *E. coli* collected in a lagoon contained a chromosomally-encoded OXA-48. Resfinder identified an average of 5 other antimicrobial resistance genes on carbapenemase plasmids, the most common of which conferred resistance to sulphonamides, tetracycline, trimethoprim and aminoglycosides. Genotypic analysis is ongoing and when coupled with the phenotypic and geographical data may reveal further insights into the ecology, evolution and dissemination of carbapenemase producing strains, carbapenemase genes and mobile genetic elements in Galway from a One Health perspective.



Conclusions

The training and harmonisation elements of the STM were successful, resulting in successful training of Dr Burke in nanopore sequencing and hybrid sequence analysis methodology and transfer of the protocol to his home Institution. The research outcome of the STM was also highly successful, resulting in 39 complete hybrid-assembled CPE genomes including 38 fully circularised CPE plasmids. Further analysis will result in a high quality scientific publication. Furthermore, the STM has strengthened links between NUI Galway's ARME lab and UCM's ARU lab, paving the way for further One Health collaborative research on antimicrobial resistance.



List of dissemination and communication activities

Not applicable

Scientific outputs

A scientific publication on the findings of this project is expected to be produced in the first half of 2022.

Testimonial

This Short Term Mission allowed me to learn cutting edge techniques in bacterial sequence analysis, which will greatly benefit researchers within my lab and our collaborators in Ireland's National CPE reference lab. I had an amazing experience in Madrid - thanks to Bruno, Carlos and everyone at the ARU for making me feel like one of the team! This mission has paved the way for further scientific collaboration and intercultural opportunities for our lab members. Thanks to the One Health EJP for making this mission possible.



2.2 Short Term Mission 1 Case Study



Image: Flickr

SHORT TERM MISSIONS

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- Sharing scientific expertise, methodologies, equipment and facilities to harmonise the existing approaches and methodologies within the large
- OHEJP European network Driving the research forward in a collaborative and non-duplicative fashion to strengthen both the scientific capacity within the OHEJP
- Contributing to the future prevention, preparedness, detection and response of the EU to foodborne and other emerging threats across human-animal-environmental sectors.

CarbaPlasmid – Tracking endemic carbapenemase plasmids in human, animal and environmental isolates



Theme: One Health, AMR
Home Institute: [NUI Galway](#), Ireland
Mission Hosting Institute: [VISAVET](#), Universidad Complutense Madrid, Spain
Duration of Mission: 1 month



This STM allowed me to learn cutting edge techniques in bacterial sequence analysis, which will greatly benefit researchers within my lab and our collaborators. I had an amazing experience in Madrid: everyone at the ARU made me feel like one of the team! This mission has paved the way for further scientific collaboration and intercultural opportunities..."

Liam Burke,
NUI Galway

The aim of this mission was to develop skills in nanopore sequencing and hybrid sequence analysis in order to characterise antimicrobial resistance plasmids for One Health epidemiological investigations.

In this project, Carbapenemase-producing Enterobacterales (CPE) isolated from the natural environment, hospital wastewater, the hospital environment and hospital patients in Galway, Ireland, were analysed by hybrid sequencing analysis. Several techniques and applications for analysis of hybrid bacterial sequence data were learned. A harmonised hybrid sequence analysis pipeline was successfully transferred between One Health EJP partners Universidad Complutense Madrid (UCA) and NUI Galway.

This short term mission developed capacity for long-read sequencing and hybrid sequence assembly in Irish OHEJP partner NUI Galway, which will also be used to support the surveillance function of the Irish National CPE Reference Laboratory. This expertise will be fundamental in ongoing and future collaborative AMR research projects with OHEJP partners and in the surveillance of endemic carbapenemase plasmids in a One-Health context in Ireland.

The data will contribute to bridging the knowledge gaps that exist in our understanding of the role of the environment in the persistence, evolution and transmission of AMR plasmids and the propensity for carbapenemase plasmid transfer between different bacterial species and strains in distinct ecological niches. It may also identify emerging AMR threats from a One Health perspective.

The training and harmonisation elements of the STM were successful, resulting in successful training of Dr Burke in nanopore sequencing and hybrid sequence analysis methodology and transfer of the protocol to his home Institution. The research outcome of the STM was also highly successful, resulting in 39 complete hybrid-assembled CPE genomes including 38 fully circularised CPE plasmids. Further analysis will result in a high quality scientific publication. Furthermore, the STM has strengthened links between NUI Galway's ARME lab and UCM's ARU lab, paving the way for further One Health collaborative research on antimicrobial resistance.

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Figure 2: This case study can be found on the OHEJP website: <https://onehealth.ejp.eu/short-term-missions-2021/>



3. Short Term Mission 3

[Zoonotic pathogen detection in rats](#)

The third STM funded for 2021 was awarded to Marieke de Cock from the National Institute of Public Health and the Environment (RIVM) in the Netherlands to visit Friedrich-Loeffler-Institut (FLI) in Germany for 8 weeks. This STM aligns with the following thematic areas: One Health Missions- Veterinary, Food, Medical and or Environmental research, harmonisation of diagnostics tests, platforms and research. This STM mission has been postponed to 2022 due to the COVID-19 pandemic.

4. Short Term Mission 4

[Developing the TELE- Vir bioinformatics toolkit for rapid characterization of emerging threats: an intersectoral exchange](#)

The fourth STM funded for 2021 was awarded to Carlijn Bogaardt, from the University of Surrey in the UK to visit the Instituto Nacional de Saúde Doutor Ricardo Jorge (INSA) in Portugal for 4 weeks. This STM aligns with the following thematic area: Integration of microbial, risk assessment and surveillance activities. This mission was expected to take place in 2021, however this wasn't possible due to COVID-19 restrictions. Carlijn has now left the University of Surrey, therefore this mission has been cancelled.