

# Introduction of WGS for *Salmonella* and *Campylobacter* at Mater Dei Hospital, Malta

Dr Rodianne Abela

Dr Graziella Zahra

(Microbiology Laboratory)

# Introduction

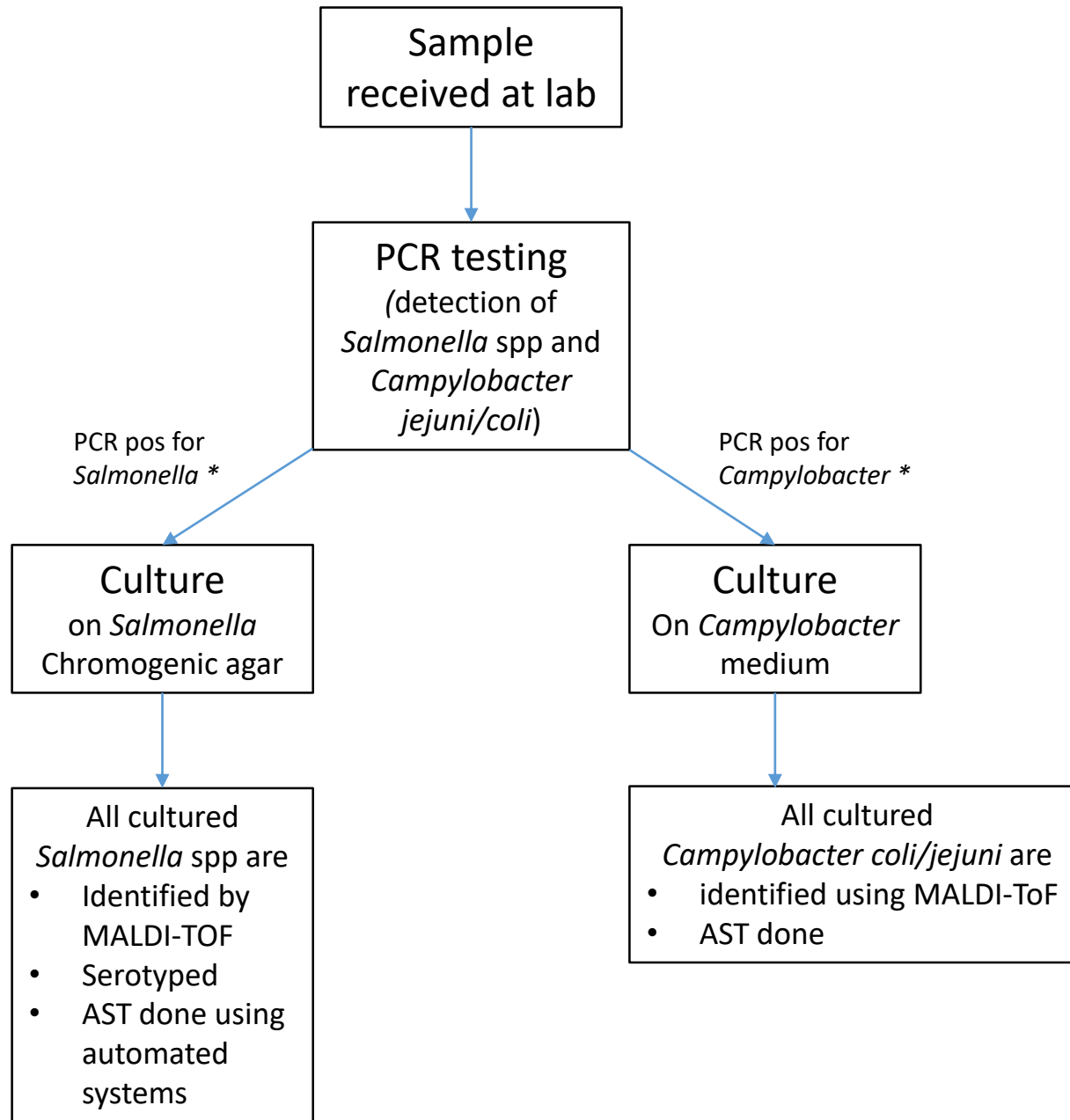
- The Maltese Archipelago is one of the smallest in the world.
- 316sq km
- Also one of the most densely populated, with over 518,000 residents.

# Background

- Laboratory located at Mater Dei Hospital
- the only state funded clinical microbiology laboratory
  - is primarily a clinical laboratory.
  - is considered to be the reference laboratory for practically all human pathogens.
- Routinely receives samples from all the public hospitals as well as from primary care and long-term care facilities.
- Cases of *Salmonella* and *Campylobacter* infections are notifiable in Malta.

# Number of requests

	2019	2020	2021
Number of faeces samples received on a yearly basis	6348	5249	6655
Number of positive cases of <i>Salmonella</i> by PCR	169	215	265
Number of positive cases of <i>Salmonella</i> by culture	130	172	210
Number of <i>Campylobacter</i> positive cases by PCR	372	312	427
Number of positive cases of <i>Campylobacter</i> by culture	257	189	254



\* *Clinical microbiologist informed about all positive PCR results. Caring clinician informed and clinical advice given. They are also reminded about the mandatory notification to public health.*

# Culture media/ techniques

The media and enrichment broths (prepared in-house) used are as follows:

## **Campylobacter**

- *Campylobacter* Medium - BioLab<sup>®</sup> Blood Agar Base + haemolysed Horse Blood (100ml per 1.5ltrs of medium for 5-7% defibrinated horse blood) + Skirrow supplement by Abtek<sup>®</sup> and Growth Supplement for *Campylobacter* by Abtek<sup>®</sup>
- *Campylobacter* Broth - BioLab<sup>®</sup> Nutrient Broth + haemolysed Horse Blood (100ml per 1 ltr of broth for 5-7% defibrinated horse blood) + Preston supplement by Abtek<sup>®</sup> and Growth Supplement for *Campylobacter* by Abtek<sup>®</sup>

## **Salmonella**

- *Salmonella* Chromogenic Agar by Oxoid<sup>®</sup> and Selective Supplement by Chromagar<sup>®</sup>
- Selenite Broth Base by Oxoid<sup>®</sup> and Sodium biselenite
- Craige tubes for phase switching for serotyping

# Background

- Malta was offered additional support as a priority country for the FWD-AMR RefLabCap project in 2021.
- We are working closely with Egle Kudirkiene (SSI) and Rene Hendriksen (DTU) as our mentors.
- Visited the laboratory physically in June 2022.
- Regular meetings are held online to assess progress and discuss any issues that arise.

# Local situation prior to initiation the project

- All faecal samples received at the Bacteriology laboratory are tested for *Salmonella* spp and *Campylobacter coli/jejuni* by PCR and all the PCR positive samples are sent for culture and antimicrobial susceptibility testing.
- Results are available to Infectious Disease Prevention and Control Unit (IDCU, Public Health Department).
- No formal regular communication between Public Health Laboratory (performing food testing), Veterinary Services, IDCU and Microbiology Laboratory



# Local situation prior to initiation the project

- Sequencers were available at the Molecular Diagnostics section procured through different tenders including the HERA project.
  - 2 illumina Miseq
  - 1 illumina Miniseq
  - 1 illumina iSeq
- NGS was introduced in 2015 for the following:
  - HIV genotyping and resistance testing
  - HCV genotyping and resistance testing
  - 16s and 18 ITS (internal transcribed spacer) assays
  - Influenza A / B

# Improvements in service provision since the initiation of the project

- Staff members at Molecular Diagnostics-Infectious Diseases laboratory have attended training courses organised at DTU.
- The laboratory signed up for the EQA schemes organised by DTU in 2022 and 2023.

# Improvements in service provision since the initiation of the project

- As of 1st October 2022, WGS is performed on:
  - all the human *Salmonella* and *Campylobacter* isolates
  - human faeces samples that are PCR positive but fail to grow the pathogen
- A WGS run is performed on a weekly basis and results are communicated to the IDCU.
- Bioinformatics is performed using available pipelines:
  - Enterobase
  - PubMLST
  - AMR: Resfinder, Point finder

# Improvements in service provision since the initiation of the project

- Since January 2023, regular meetings are being held between microbiologists, public health consultants, Microbiology laboratory management, veterinary department and public health laboratory management.
- During these meetings, we have agreed to start sequencing *Salmonella* and *Campylobacter* isolates from veterinary and food lab that have been identified since October 2022.
- We have also sequenced MDR *Salmonella* Haifa isolates that were confirmed as a cluster. All *Salmonella* Haifa isolates identified at veterinary laboratory were also submitted for testing.
  - No link was established between human and veterinary isolates.

# Ongoing Challenges

- Human resources and training
- Antibiotic susceptibility testing as per harmonised protocol
  - Do we need phenotypic testing for all antibiotics if we're doing genotypic testing?
- Space limitations in the laboratory
- No dedicated bioinformatician available
- IT infrastructure to allow easier access to data from the different departments

# Acknowledgements

- All Bacteriology scientific staff involved in this project.
- All Molecular Diagnostics – Infectious Diseases laboratory involved in this project
  
- Dr Christopher Barbara
- Dr Claire Marantidis Cordina
- Ms Julie Haider
- Mr Robert Cassar
  
- Dr Tanya Melillo
- Dr Maria- Louise Borg
- National Veterinary Laboratory
- National Public Health Laboratory