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National *Clostridioides difficile* Reference Laboratory Service provided by, PHL, HSE, Dublin. Annual Report 2022

Summary

- 406 specimens were received from 388 patients
- 381 *C. difficile* isolates/406 (93.8 %) samples characterized:
  - 336/381 (88.2 %) were from *C. difficile* PCR positive stools
  - 45/381 (11.8 %) were *C. difficile* isolates submitted directly to the NRL
- Samples were submitted from 28 participating laboratories.
- Of the 381 isolates (93.8%) were confirmed as toxigenic *C. difficile* culture positive
  - 100% were *tcd*A & *tcd*B PCR positive
  - For stool samples received, 324/336 (96%) were GDH EIA positive and 223/336 (66.3%) were toxin A/B EIA positive.
  - Of the 336 stool samples, 324/336 (96%) were GDH EIA positive.
  - Of the 379 isolates tested for vancomycin & metronidazole phenotypic resistance, 100% were susceptible (EUCAST 2022 criteria)
- 381 isolates /381 passed WGS QC for analysis
  - Majority of isolates were toxigenic producing *C. difficile*.
  - 374/381 (98.2%) *C. difficile* isolates had *tcd*A,
  - 377/381 had *tcd*B (98.9%) genes detected
  - 19% isolates having binary *cdt*A & *cdt*B genes encoding enhanced virulence detected by WGS.
- Sequence type ST11, 60/381 (16%) was predominant among *C. difficile* nationally
- 45 different sequence types were detected nationally
- Whole Genomic Sequencing (WGS) identified 68 potential clusters for client alerts in 2022 warranting consideration of further epidemiology investigations.

Introduction

This is the first annual report for the newly established Irish National *Clostridioides difficile* Laboratory services (NRL). The *Clostridioides difficile* NRL was awarded to Public Health Laboratory (PHL), HSE, Dublin in September 2021 following a competitive national tender. Funding was provided to process 1000 *C. difficile* isolates annually as part of this process. Service commenced in July 2022 following communication with all 60 hospitals participating in the National Enchanced CDI Surveillance (as notified by case definitions in Appendix 1) and their local microbiology laboratories (see appendix 2 & 3).

The delay between the awarding of tender and service commencement was due primarily to connectivity issues between the HSE network and a 2nd Illumina MiSeq which was used for whole genome sequencing (WGS) following the HSE cyber-attack that occurred in May 2021. A proposed solution was agreed in May 2022 but full functionality of the 2nd Illumina MiSeq only returned in February 2023. A summary of the timeline since awarding of tender is detailed in Appendix 4.

Specimen submission

Participating laboratories and clients are advised to send stool samples that are positive for *C. difficile* from (a) PCR or (b) GDH and toxin testing. Clients are encouraged to send isolates should they have capacity to culture *C. difficile* from stools in their laboratories.

- A total of 406 specimens were received from 388 patients.
- 381 samples that were dated from Q4 2021 and 2022 were successfully cultured and underwent WGS.
    - 336/381 (88.2%) were recovered from stools.
    - 45/381 (11.8%) were recovered from isolates submitted.

A total of 28 laboratories from across the country submitted samples to the service.

Culture and phenotypic methods

The submitted specimens were processed as follows:

1. When a faeces sample was submitted, an alcohol shock method was applied to isolate *C. difficile*.
2. Following this step, the inoculum or submitted isolate was placed on *C. difficile* selective agar and Brazier's CCEY and incubated anaerobically at 35°C for 48 hours.
3. PCR was performed on suspect colonies which were then subbed to fastidious anaerobic agar and chromeID® C.*difficile* agar, and incubated anaerobically for 48 hours at 36°C.
4. Phenotypic antimicrobial susceptibility testing (AST) was set up on fastidious anaerobic agar from the purity plates using minimum inhibitory concentration method by ETEST® strips.
5. Confirmed isolates are stored on beads and referred for whole genome sequencing.
6. If original culture failed to yield viable *C. difficile* isolates after step 2, an additional step involving inoculating sample into cooked meat broth was carried out and sample processing continued from step 2.

Phenotypic antimicrobial susceptibility results

AST by minimum inhibitory concentration (MIC) was performed according to EUCAST guidelines on successfully recovered *C. difficile* isolates. Interpretation of MICs is based on EUCAST breakpoints for *C. difficile*. 379 isolates had completed phenotypic testing during this time period. All isolates were susceptible to metronidazole and vancomycin. At the time of writing, fidaxomicin susceptibility testing is not available due to a lack of standardised methods but the service is exploring options for testing in 2023.

Whole genome sequence characterization

A total of 381 *C. difficile* isolates successfully completed WGS. High-quality DNA was extracted from confirmed isolates and DNA libraries were prepared using the Illumina DNA Prep kits and sequenced on an Illumina MiSeq instrument. Sequence yields that passed quality parameters (Q-score, GC content yield, coverage) were assembled *de novo* using the BioNumerics platform (version 8.1.1). These genome assemblies were then assessed for quality using the metrics N50, contig length, total sequence length, and percent core coverage. WGS analysis for sequence types (ST), virulence determinants and cluster detection was completed for the 381 isolates that passed the quality criteria.

45 different *C. difficile* sequence types (ST) were confirmed, demonstrating significant heterogeneity in the current national *C. difficile* collection. 6 STs were predominant, each with greater than 17 isolates. While ribotyping is not performed at *C. difficile* NRL, there is some broad correlation between ribotypes (RT) & WGS derived ST. For example, **ST11** (16%) may equate to RT O78, **ST8** (13%) – RT O02, **ST2** (12%) to RTs O14, O20, O76, 220,**ST6** (7%) – RT O05, **ST10**(6%)-RT O15 & **ST3**(4.5%) – RTs O01, O09, O72, 115..

> **Please note ribotyping is not part of the Irish NRL *C. difficile* service, as WGS gives more comprehensive data (sequence types, virulence factors & genomic AMR determinants) and is the preferred methodology employed in this NRL.**

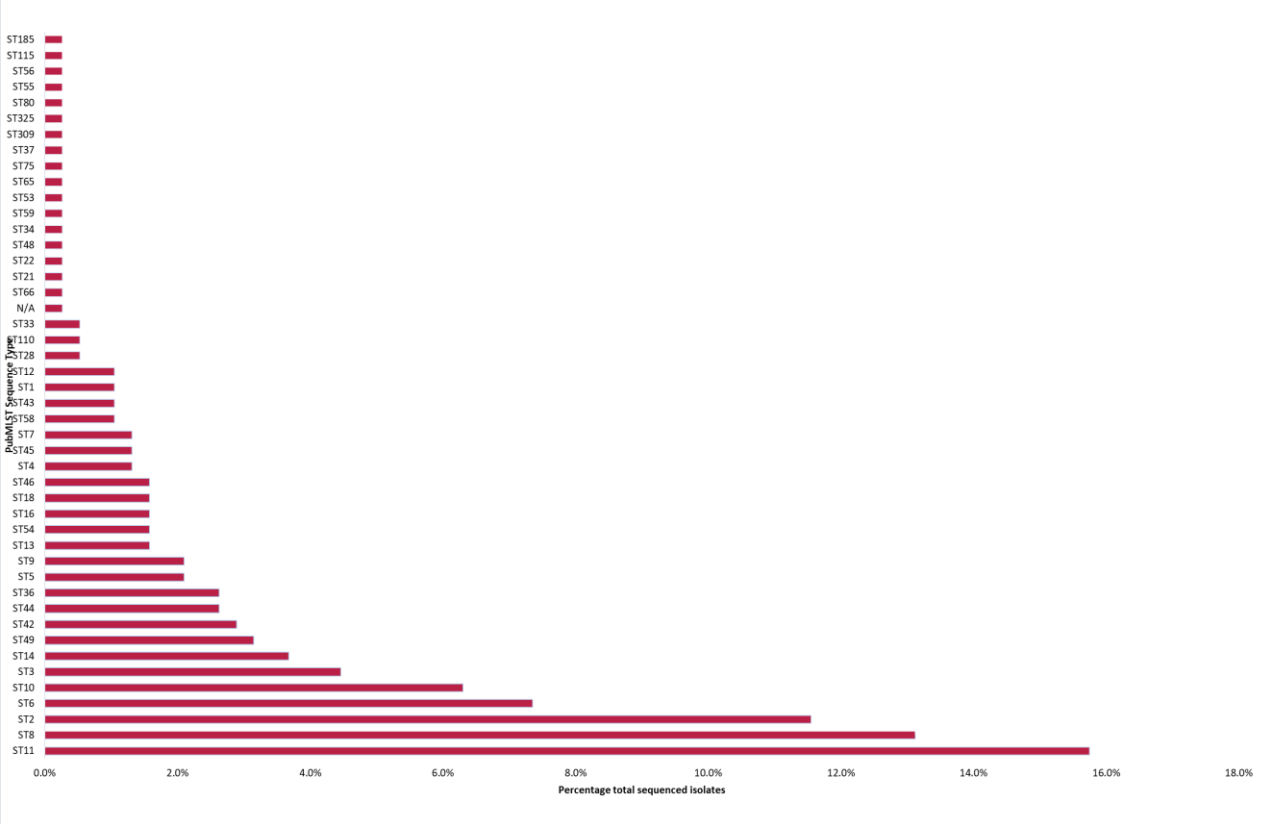

Figure 1 NRL *C. difficile* Sequence types 2021Q4 -2022, n= 381

Virulence factors

Table 1 describes the breakdown of virulence factors found in isolates submitted to the service (n=213). Of 381 isolates, all isolates were toxigenic producing *C. difficile* isolates. 377 (97.6%) isolates had both *tcdA* and *tcdB* present. Only 4 isolates had *tcdA* present but *tcdB* absent. Of the 4 samples, 2 were submitted from laboratories that used GDH as the first step of *C. difficile* testing. One submitted stool sample had 2 distinct *C. difficile* isolates, one having both *tcdA* and *tcdB* present and one with *tcdA* only.

The binary toxin (*cdt*A & *cdt*B) which is associated with enhanced virulence was found in all ST11 *C. difficile* isolates (n=60).

	Total cases		ST11		ST8		ST2		ST6		ST10		ST3	
	n	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**
Total CDI cases sequenced	381	-	60	16%	50	13%	44	12%	28	100%	24	-	17	4%
CDI toxin genotype														
tcdA positive	374	98%	56	93%	50	100%	44	100%	28	100%	24	100%	17	100%
tcdB positive	377	99%	59	98%	49	98%	44	100%	28	100%	23	96%	17	100%
tcdC positive	313	82%	0	0%	50	100%	44	100%	28	100%	24	100%	17	100%
cdtA/cdtB positive	73	19%	60	100%	0	0%	0	0%	0	0%	0	0%	0	0%
CDI isolates identified as part of clusters	197	52%	50	83%	31	62%	12	27%	12	43%	14	58%	14	82%

Table 1 C. difficile NRL WGS profile of all referred C. difficile isolates, 2022 (n=381)

Cluster analysis

Analysis was performed weekly by cgMLST to detect allelic differences (AD). If zero AD, they were considered 'Genetically indistinguishable', a ≤2 allele difference may be considered 'Very Closely Related'. Such WGS cluster reports were issued to the relevant clients. In addition surveillance notes were also sent to clients if AD ≤5 were detected for future monitoring purposes.

The following is a summary of clusters that were identified through WGS cluster analysis. Individual cluster reports were sent to hospital sender(s), relevant public health departments and the Health Protection and Surveillance Centre (HPSC) at the time of identification:

68 WGS cluster reports and 2 WGS surveillance notes were issued in 2022

- 2 Surveillance notes of ≤5 allele difference (AD) involved 2 separate groupings of ST11, the predominant clonal ST in Ireland
    - ST11 with 20 isolates sent from 15 Hospital laboratories
    - ST11 with 8 isolates sent from 3 Hospital laboratories

Of the 68 Clusters with ≤2 (AD):

The 8 largest Clusters issued with ≤2 (AD) ranged with 6-9 isolates in each report

- 1 ST11 Cluster with 9 isolates, involved 4 hospitals
- 1 ST11 Cluster with 8 isolates, involved 7 hospitals
- 3 Clusters had 7 isolates each, ST36 Cluster involved 5 hospitals, ST11 Cluster involved 6 hospitals & ST3 Cluster involved 6 hospitals
- 2 ST8 Clusters had 6 isolates each, one involved 4 hospitals and the other involved 2 hospitals
- 1 ST11 Cluster had 5 isolates that involved one hospital.

Of the remaining 60 Clusters that were issued, the breakdown was the following:

- 5 clusters had four isolates each
- 10 clusters had three isolates each
- 45 clusters had two isolates each

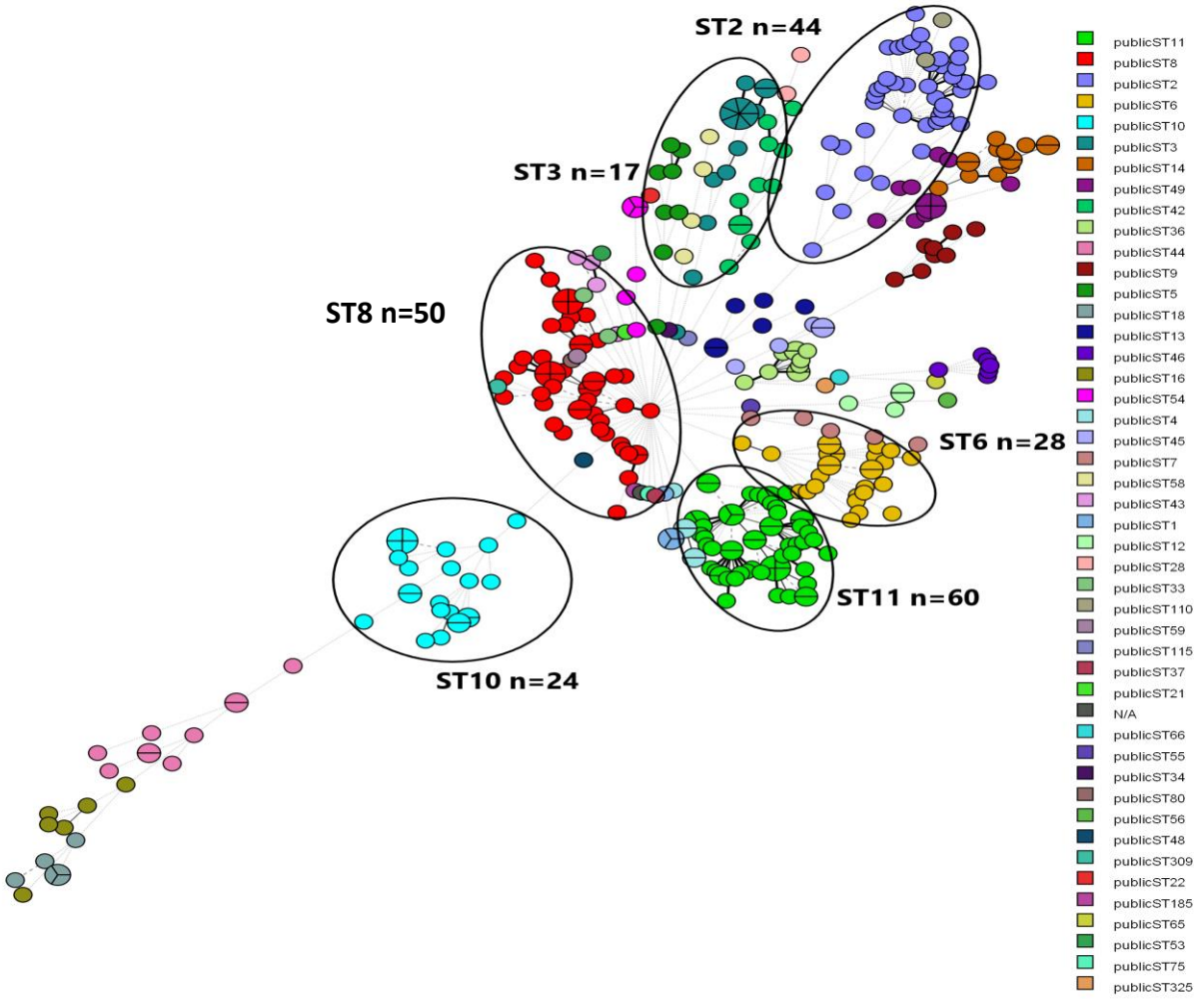

Figure 2 Minimum spanning tree of *C. difficile* cgMLST Sequence types in 2022 (n= 381)

The minimum spanning tree in figure 2 demonstrates the grouping of the predominant sequence types by coloured circles.

External Stakeholder Engagement

Reports

Appendix 5 describes all the reports issued as part of the *C. difficile* NRL.

NRL-HPSC surveillance meetings

Routine engagement with Health Protection Surveillance Centre (HPSC) occurred to address a variety of joint surveillance requirements, data sharing that is in line with GDPR legislation, NRL report distribution lists, the frequency and contents of NRL reports. The first combined HPSC-NRL *C. difficile* quarterly report was published in November 2022 as part of the Quarter 2 2022 National report.

These reports are available from the HPSC websites. Available at: https://www.hpsc.ie/a-z/microbiologyantimicrobialresistance/clostridioidesdifficile/enhancedsurveillance/quarterlyreports/

Reports to AMRIC by NRL *C. difficile*

Two reports were issued to AMRIC as requested in June 2022 & Feb. 2023. These included an update on the progress of the new NRL *C. difficile* service in compliance with the terms of the awarded tender. The AMRIC team acknowledged the NRL achievements.

NRL Presentations

(1) 'Focus of Infection' RCPI webinar 19.1.23. 'The new *Clostridioides difficile* national reference laboratory service- first results' Dr. Eleanor McNamara, Director NRL

(2) Biomedical, March 2023. 'NRL *C. difficile*- an update on the new service' Dr Anne Carroll, Chief Medical Scientist NRL.

PHL HSE Dublin NRL open day for service users 9.12.22.

A variety of NRL clients including Public Health Doctors, Medical Scientists, and Surveillance Scientists attended a day of lectures & discussion on the new NRL service. It also included a tour of the NRL. The feedback was very positive.

International linkages

The Irish *C. difficile* NRL has links with:

- ECDC sponsored HCAI network
- ECDC coordinated *C. difficile* WGS EQA
- ESCMID CDI study group
- The Irish CDI Network (ICDI-Net)
- UK *C. difficile* Reference Laboratory (as part of the UK Anaerobic Reference Laboratory, Cardiff, Wales)

Other Activities

Research

A research project undertaken by Dr Leike Brouwer, ECDC EUPHEM Fellow Cohort 2022 is ongoing on the comparability of phenotypic to genomic determinants of antimicrobial resistance (AMR) in *C. difficile*. It is hoped this will influence the future NRL service to replace phenotypic antimicrobial sensitivity with validated WGS detection of a broad range of genomic determinants of AMR, which would be more beneficial to clients. Clients will be updated in 2023 on these developments.

Service Development 2023 and beyond

- **Demand management within tender contract**
  - Consideration to address samples prioritization if submitted sample numbers are in excess of the annual tender contract
- **Phenotypic AMS testing moves from comprehensive to sentinel testing**
  - As no phenotypic resistance was detected for vancomycin & metronidazole in any 2022 *C. difficile* isolates, it was decided to reduce this to sentinel phenotypic AMR testing (25% coverage) for 2023. This will speed up workflow & save costs, while validation of WGS AMR determinants is ongoing.
  - Progression of fidaxomicin phenotypic antimicrobial susceptibility testing.
- **Deepening working relationships with relevant clients to strengthen epidemiological investigation of reported clusters found through WGS**.
  - Currently there is no formalised system in place to document nationally the results of local CDI cluster investigations based on NRL genomic data. This will improve as multi-disciplinary groups such as 'The Irish *C. difficile* Infection Network (ICDI-Net)'evolve. ICDI-Net had its first meeting in March 2023.

Director retirement & replacement

Dr. Eleanor McNamara, Director has retired in June 2023. Dr Tee Keat Teoh is the newly appointed Director & Consultant Microbiologist of the PHL incorporating NRL services. Dr. Brian O'Connell & Prof Johannes Wagener continue to also provide Consultant PHL & NRL services. Please contact PHL for the Consultant on duty & for any NRL queries (phone 01-755214 or email phl.dublin@hse.ie).

Summary

The PHL HSE Dublin staff have shown tremendous dedication to the development of this new Irish NRL *C. difficile* service. This report documents the completed first year of service. Client feedback has been positive with many suggestions for future developments welcomed. Significant progress on the uptake of this national service is occurring as more laboratories become familiar with the benefits of this Irish service. Excellent inter- agency co-operation with HPSC is on-going and further collaborative protocols will be progressed in 2023. We do

encourage those laboratories not currently submitting isolates to the NRL to do so in 2023 to strengthen the human epidemiological data of *C. difficile* in Ireland.

Acknowledgments

The National Reference Laboratory Service for *C. difficile* would like to sincerely thank all who have contributed to the development of the service. We would like to give special thanks to colleagues in the UK Anaerobic Reference Unit, Cardiff, Wales for sharing their knowledge to help set up the methodology in our laboratory. We would like to mention the many colleagues from the ECDC, HPSC, AMRIC, Microbiology surveillance scientists, Infection Prevention and Control Nurses, Microbiology laboratory scientists, Clinical Microbiologists, along with all the staff of the Departments of Public Health across Ireland in supporting the work that has been completed to date.

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Appendices

1. CDI Case definitions
2. NRL *C. difficile* Letter to clients June 2022
3. National CDI Enhanced surveillance participating Hospital
4. Appendix 4 Timeline of establishment of regular service for the C.diff NRL following awarding of tender.
5. List of regular C. difficile NRL reports issued:

Appendix 1
Case Definitions for Surveillance of *Clostridioides difficile* Infection

***For surveillance purposes, a confirmed Clostridioides difficile* infection (CDI) case is a patient two years or older, to whom one or more of the following criteria applies:**

- Diarrhoeal* stools or toxic megacolon, with either a positive laboratory assay for *C. difficile* toxin A (TcdA) and/or toxin B (TcdB) in stools or a toxin-producing *C. difficile* organism detected in stool via culture or other means.
- Pseudomembranous colitis (PMC) revealed by lower gastrointestinal endoscopy.
- Colonic histopathology characteristic of *C. difficile* infection (with or without diarrhoea) on a specimen obtained during endoscopy, colectomy or autopsy.

*** Diarrhoea is defined as three or more loose/watery bowel movements (which are unusual or different for the patient) in a 24 hour period**

CASE TYPE

- **New Case of CDI:**
  - The first episode of CDI, **OR**
  - A subsequent episode of CDI with onset of symptoms **more than eight weeks** after the onset of a previous episode.

- **Recurrent Case of CDI:**
  - A patient with an episode of CDI that occurs **within eight weeks** following the onset of a previous episode **provided that CDI symptoms from the earlier episode resolved with or without therapy.**

ONSET

- **Healthcare onset** » Symptoms start during a stay in a healthcare facility.
- **Community onset** » Symptoms start in a community setting, outside healthcare facilities.
- **No information available** » If no information was available on onset of symptoms

ORIGIN

- **Healthcare-associated case.** This is a CDI patient with either:
  - Onset of symptoms at least 48 hours following admission to a healthcare facility (healthcare-onset, healthcare-associated), OR
  - With onset of symptoms in the community within four weeks following discharge from a healthcare facility (community-onset, healthcare-associated).
- **Community-associated case.** This is a CDI patient with either:
  - Onset of symptoms while outside a healthcare facility, and without discharge from a healthcare facility within the previous 12 weeks (community-onset, community-associated), OR
  - With onset of symptoms within 48 hours following admission to a healthcare facility without residence in a healthcare facility within the previous 12 weeks (healthcare-onset, community-associated).
- **Discharged 4 – 12 weeks from a healthcare facility**

»This is a CDI patient who was discharged from a healthcare facility between four and 12 weeks before the onset of symptoms.

- **No information available**

SEVERE CDI Case

This is a CDI patient to whom any of the following criteria apply:

- Admission to an intensive care unit for treatment of CDI or its complications (e.g., for shock requiring vasopressor therapy)
- Surgery (colectomy) for toxic megacolon, perforation or refractory colitis
- Death within 30 days after diagnosis if CDI is either the primary or a contributive cause

Appendix 2: NRL *C. difficile* Letter to clients July 2022

19th July 2022

Re: **Update on National Reference Laboratory (NRL) *Clostridioides difficile* services at PHL HSE Dublin**

Dear Colleague

We are pleased to inform you that the new NRL *C. difficile* services at PHL HSE Dublin has commenced. This is later than we had hoped due to delays mostly outside of our control e.g. appointment of new personnel, installation of equipment along with method validation & verification including excellent intra-laboratory comparative WGS studies. However while we are contracted to process 1000 samples PA, we currently still do not have our 2nd *C. difficile* dedicated sequencer networked which is being addressed by 'Illumina' & HSE IT. Thus we will have to restrict our service to prioritised samples until further notice.

We will now accept (A) Isolates of *C. difficile* (preferential) or (B) *C. difficile* PCR positive stools for further culture & molecular characterisation. All samples need to be submitted using our NRL *C. difficile* request form https://www.hse.ie/eng/services/list/5/publichealth/publichealthlabs/public-health-laboratory-dublin/request-forms.html

Analysis will include;

- **Toxigenic culture** with phenotypic sensitivity testing to Vancomycin & Metronidazole.
- **Genomic analysis** for;
    - Speciation
    - Virulence toxin genes – *tcd*A, *tcdB, tcdC, tcdR, tcdE* & *cdtA*, *cdtB*,
    - Sequence type
    - core genome multilocus sequence type (cgMLST) WGS cluster analysis

Reports issued from NRL will be;

- Individual culture & AMS Reports to be sent contemporaneously to the sender
- Weekly review of the *C. difficile* WGS database for clusters will occur & if clusters are identified a report will be sent to the sender, relevant Departments of Public Health & HPSC
- Monthly anonymised national cumulative *C. difficile* Excel tables will be issued to;
    - All consented senders hospitals, (each hospital will be given a unique code sent earlier)

    - The data included will be NRL sample no, sample date, NRL sample receipt date, hospital code, toxigenic culture result, phenotypic AMS , Virulence toxin genes & sequence type
- Quarterly combined NRL-HPSC enhanced *C. difficile* surveillance reports are proposed, with details to follow.
- Annual NRL *C. difficile* report

Due to our limited capacity, we will currently prioritise *C. difficile* outbreak samples & severe *C .difficile* infection (CDI) cases for urgent investigation with surveillance & routine samples fulfilling the remaining capacity. Ensure such priority samples are documented on the request form & are phoned or e-mailed to (phl.dublin@hse.ie) in advance to the NRL.

Generally only send 1 sample /case, see national guidelines (https://www.hpsc.ie/az/microbiologyantimicrobialresistance/infectioncontrolandhai/guidelines/File,13950,en.pdf) unless recurrent or reinfection with *C. difficile* is considered.

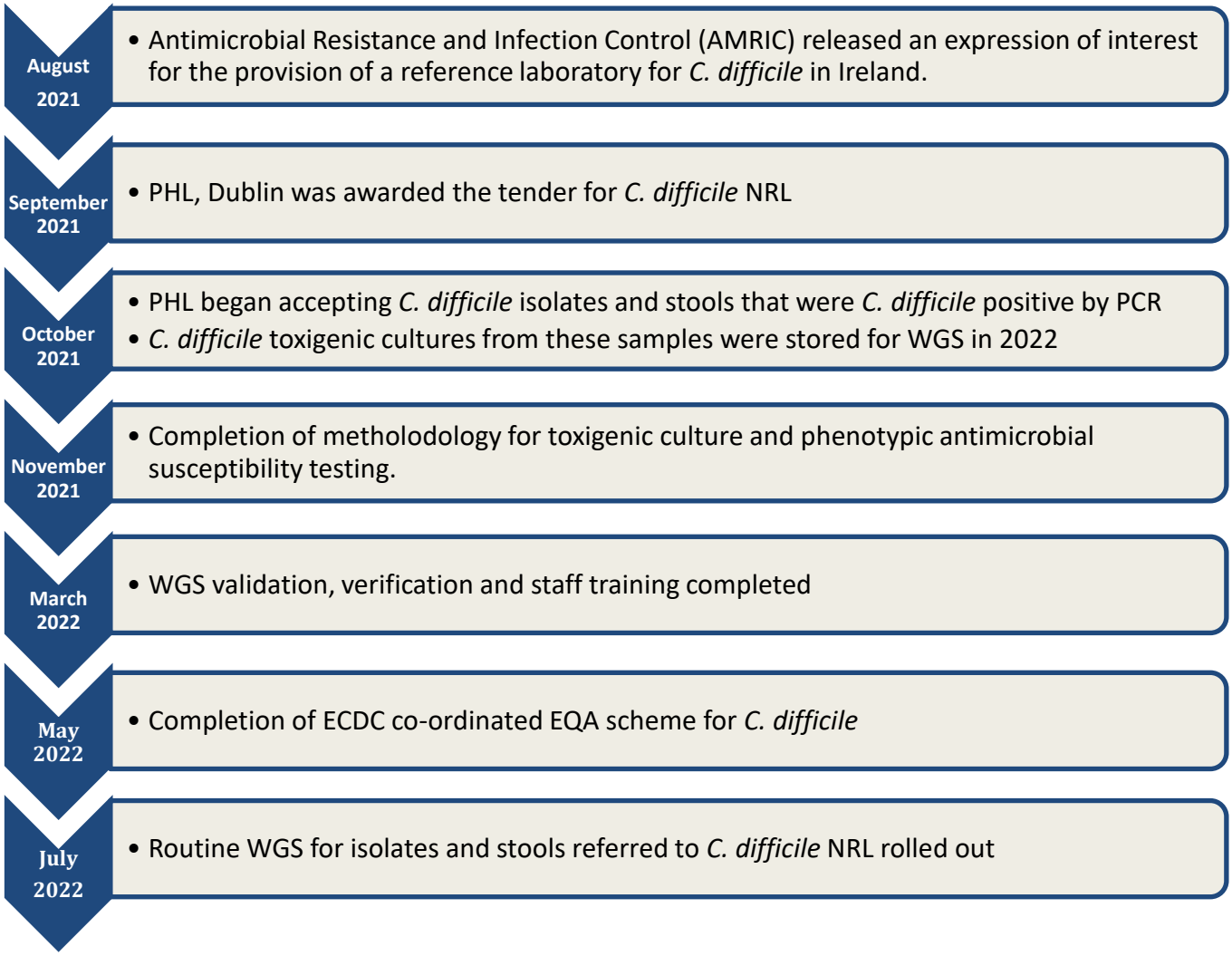
This is an exciting new national service development and we encourage any queries or feedback on the service to be e mailed at 'phl.dublin@hse.ie' (which is monitored during workdays) or phoned directly to PHL Dublin at 017955175.

Yours sincerely

Appendix 3: National CDI Enhanced surveillance participating Hospitals (source HPSC)

Hospital Group	Hospital Name	Category	Type of Hospital	Area
Dublin Midlands	Coombe Women and Infant's University Hospital	Specialist	-	B
	Midland Regional Hospital Portlaoise	General	Model 3	B
	Midland Regional Hospital Tullamore	General	Model 3	B
	Naas General Hospital	General	Model 3	B
	St James's Hospital	Tertiary	Model 4	B
	St Luke's Hospital, Dublin	Specialist	-	B
	Tallaght University Hospital	Tertiary	Model 4	B
Ireland East Hospital Group	Cappagh National Orthopaedic Hospital, Dublin	Specialist	-	A
	Mater Misericordiae University Hospital	Tertiary	Model 4	A
	Midland Regional Hospital Mullingar	General	Model 3	B
	National Maternity Hospital, Holles Street	Specialist	-	C
	National Rehabilitation Hospital, Dun Laoghaire	Specialist	-	C
	Our Lady's Hospital, Navan	General	Model 3	A
	Royal Victoria Eye & Ear Hospital, Dublin	Specialist	-	C
	St Columcille's Hospital, Loughlinstown	General	Model 2	C
	St Luke's General Hospital, Kilkenny	General	Model 3	C
	St Michael's Hospital, Dun Laoghaire	General	Model 2	C
	St Vincent's University Hospital	Tertiary	Model 4	C
	Wexford General Hospital	General	Model 3	C
RCSI Hospital Group	Beaumont Hospital	Tertiary	Model 4	A
	Cavan General Hospital	General	Model 3	A
	Connolly Hospital, Blanchardstown	General	Model 3	A
	Louth County Hospital, Dundalk	General	Model 2	A
	Our Lady of Lourdes Hospital, Drogheda	General	Model 3	A
Saolta Hospital Group	Letterkenny University Hospital	General	Model 3	F
	Mayo University Hospital	General	Model 3	F
	Portiuncula University Hospital	General	Model 3	F
	Roscommon University Hospital	General	Model 2	F
	Sligo University Hospital	General	Model 3	F
	University Hospital Galway	Tertiary	Model 4	F
South/South West Hospital Group	Bantry General Hospital	General	Model 2	D
	Cork University Hospital	Tertiary	Model 4	D
	Cork University Maternity Hospital	Specialist	-	D
	University Hospital Kerry	General	Model 3	D
	Lourdes Orthopaedic Hospital, Kilcreene, Kilkenny	Specialist	-	C
	Mallow General Hospital	General	Model 2	D
	Mercy University Hospital, Cork	General	Model 3	D
	South Infirmary - Victoria University Hospital, Cork	General	Model 2	D
	South Tipperary General Hospital, Clonmel	General	Model 3	C
	University Hospital Waterford	Tertiary	Model 4	C
UL Hospital Group	Croom Hospital	Specialist	-	E
	Ennis Hospital	General	Model 2	E
	Nenagh Hospital	General	Model 2	E
	St John's Hospital	General	Model 2	E
	University Hospital Limerick	Tertiary	Model 4	E
	University Maternity Hospital Limerick	Specialist	-	E
Private Hospitals	Aut Even, Kilkenny	Private	-	
	Beacon Hospital, Dublin	Private	-	
	Blackrock Clinic	Private	-	
	Bon Secours, Cork	Private	-	
	Bon Secours, Galway	Private	-	
	Bon Secours, Glasnevin	Private	-	
	Bon Secours, Tralee	Private	-	
	Galway Clinic	Private	-	
	Hermitage Medical Clinic, Dublin	Private	-	
	Mater Private, Dublin	Private	-	
	Mater Private, Cork	Private	-	
	St Vincents Private Hospital	Private	-	
Children's Health Ireland	Children's Health Ireland at Tallaght	Specialist	-	
	Children's Health Ireland at Temple St	Specialist	-	

Appendix 4 Timeline of establishment of regular service for the C.diff NRL following awarding of tender.



Appendix 5 List of regular *C. difficile* NRL reports issued:

1. **Individual sample Reports** of *C. difficile* culture with sentinel phenotypic AMS data (if performed on that sample) is sent contemporaneously to the sender.
2. **Cluster Reports:** Weekly review of the *C. difficile* WGS database for cgMLST clusters (Allelic difference ≤2) occurs. If clusters are identified, a unique numbered cluster report is issued to the relevant:
    a. Hospital sender(s).
    b. Regional Departments of Public Health.
    c. HPSC.

    Individual referring hospital(s) can use such data to initiate relevant investigation of these clusters.

    PHL Consultants & Specialist Scientific staff can address any queries with regards to cluster report interpretation.

3. **Monthly anonymized national cumulative *C. difficile* Excel tables** are issued to:
    a. All consented senders hospitals, (each hospital has a unique code)
    b. HPSC
        - The data included is; NRL sample no, Senders sample no, sample date, NRL sample receipt date, Hospital code, toxigenic culture result, sentinel phenotypic AMS , Virulence toxin genes, sequence type & identified clusters.
        - This cumulative 'rolling' data sheet will enable each hospital laboratory to search for their own data using their unique Hospital code.
4. **Quarterly combined NRL-HPSC enhanced *C. difficile* surveillance reports** are issued. The degree of matching the HPSC & NRL databases are dependent on accurate sender's information. The first report of Q2 2022 was issued in Nov. 2022 & the second report Q3 2022 was issued in February 2023. A combined Q4 2022/Q1 2023 report is currently in progress.
5. **Annual NRL *C. difficile* reports will be issued to all stakeholders**
6. **Annual combined HPSC-NRL *C. difficile* reports will also be issued to all stakeholders.**