National Carbapenemase Producing Enterobacteriaceae (CPE) Reference Laboratory Users Guide

CURRENT VERSION AMENDMENTS

Each SOP has an individual record of amendments. The amendments for the current version are listed below.

Amendment Number/Date	Version No. Discarded	Version No. Issued	Section(s) involved	Amendment
3/22.04.17	3.0	4.0	1.0	Details added regarding Acinetobacter testing
			3.0	Details added regarding Service Level Agreement
			4.1	Updated Section to reflect changes, including accepting Acinetobacter species
			4.3	Section updated to reflect relevant changes, i.e. AST no longer performed on referred isolates and WGS on relevant isolates
			4.4	Details regarding WGS added
			9.0	Target coverage updated

Change Control: MIC019/17

1.0 The Laboratory and Outline of Services

The CPE reference laboratory, department of Microbiology, University Hospital Galway provides a clinically supported service for the detection of carbapenemase producing *Enterobacteriaceae*. Although *Acinetobacter spp.* are not *Enterobacteriaceae* we are now accepting isolates of this genus in so far as possible in response to concerns regarding meropenem resistance in this genus.

This service is offered to all medical laboratories in hospitals throughout Ireland.
The Department of Medical Microbiology at GUH participates in available external quality assurance schemes and is accredited by the Irish National Accreditation Board (INAB) to undertake testing as detailed in the Schedule bearing the Registration Number 223MT in compliance with the International Standard ISO/IEC 15189:2012 3rd Edition.

This guidance for users represents an attempt to target the available resources to deliver the service so as to maximize detection of the major CPE concerns. Given the variety of known CPE enzymes, variability of gene expression, other mechanisms that result in raised carbapenem MIC and the rapidly changing epidemiology it is acknowledged that no set of selection criteria for isolate submission can ensure that every CPE producing isolate is confirmed. The CPE reference laboratory service would like to acknowledge very helpful comments and calls from colleagues in laboratories around the country highlighting specific concerns which have been considered in revising this guidance and would appreciate if you could continue to alert the service to concerns and emerging issues.

2.0 Laboratory Policy

The Division of Clinical Microbiology is committed to providing a timely, efficient and quality diagnostic and reference laboratory service to all patients, clinicians and other users of the service. Laboratory Management is committed to maintaining and continually improving the quality system so that the requirements of ISO15189 and the Irish National Accreditation Board (INAB) are met on an ongoing basis. Through its quality management system, the Division aims at all times to ensure that the service provided by this department, as defined within the current scope of accreditation (detailed in the Schedule bearing the Registration

Number 223MT) is of the standard required to achieve maximum patient benefit as defined in ISO 15189. It is Division policy that all staff are trained and have familiarised themselves with the quality documentation to ensure they can implement all policies and procedures. The Division is committed to good professional practice and to the provision of examinations that are fit for intended use to achieve a quality of service that is compliant with their quality management system and therefore the international standard ISO 15189.

3.0 User Information

Service Level Agreement (SLA)

The request form for the CPE Reference Laboratory serves as the formal 'Service Level Agreement' between the National Carbapenemase Producing *Enterobacteriaceae* Reference Laboratory [NCPERL], Diagnostics Directorate, Galway University Hospital (GUH) and the Service user.

Contact Details

This user guide is designed to assist the Client in the use of the services we offer. Further information is available by telephoning the laboratory.

Consultant with Administrative Responsibility: Prof. Martin Cormican (091) 544146

Laboratory Contact: (091) 544570 / (091) 544429
Fax: (091) 542238
email: martin.cormican@hse.ie

Address: Carbapenemase Producing Enterobacteriaceae (CPE) Reference Laboratory
Department of Medical Microbiology
University Hospital Galway
Galway

Opening Times: Monday to Friday 9.30am - 5.30pm

4.0 Services Provided

The CPE Reference Laboratory (CPERL) performs analysis on isolates received on nutrient agar slopes. The CPERL does not test primary samples, e.g. swabs, faeces, blood or food.

4.1 Isolates to be Referred for Carbapenemase Analysis

Enterobacteriaceae

- Intermediate/Resistant to:
  - Meropenem
  - Ertapenem
  - Imipenem
- Or with an MIC to Meropenem of greater than 0.125 mg/L (isolates with an MIC of 0.125 or lower do not need to be submitted)

Acinetobacter spp.

- Intermediate/Resistant to:
  - Meropenem
  - Imipenem

- **Note:**
  - *Proteus* spp. and *Morganella* spp. are intrinsically resistant to Imipenem.
  - *Enterobacter* spp. resistant to cephalosporins and with low level resistance to ertapenem but susceptible to meropenem typically represents the combination of AmpC and impermeability rather than carbapenemase production.
  - *Pseudomonas aeruginosa:* UK guidance suggests carbapenem- EDTA synergy tests for isolates resistant to meropenem, ceftazidime and piperacillin-tazobactam although notes that false positive are common with this test. Previous experience at GUH also indicated that all of the positives detected with one commercially available EDTA-carbapenem synergy test were false positive. The National Reference Laboratory Service would not have the resources to process all such *Pseudomonas aeruginosa* isolates and the yield of CPE would be very low. However if Clinical Laboratories are concerned

about specific strains particularly if there appear to be 2 or more linked patients or recent hospitalization outside of Europe the reference laboratory will accept the isolates by prior arrangement.

- *Stenotrophomonas maltophilia* is intrinsically resistant to carbapenems.

4.2 Request Form & Labelling Requirements

The CPE request form must accompany and identify all isolates. Currently the CPE request form is available to download on the National Salmonella, Shigella and Listeria Reference Laboratory (NSSLRL) website:

http://www.nuigalway.ie/salmonella_lab/downloads/cpe_request_form.pdf

The **request form** must contain the following information:

- Patients Name
- Patients Date of Birth
- Referring laboratory name and contact details
- Laboratories isolate reference number

The referring **slope** must contain the following information:

- Patients Name and/or Date of Birth
- Laboratories isolate reference number

4.3 Tests Performed

The following tests are performed on all Query Carbapenemase producing isolates:

- Identification by MALDI-TOF
- ROSCO – phenotypic screen for Class A Carbapenemase (usually KPC), metallo-β-lactamases and OXA-48
- Molecular testing by RT-PCR for Carbapenemase genes (see Appendix 10.0 for targets covered by RT-PCR)
- Colistin MIC by broth dilution using TREK Sensititre
- WGS on carbapenemase producing *E.coli* and *K.pneumoniae* and other groups in so far as possible.

4.4 Results and Turnaround Times

The reference laboratory aims to report 95% of results within 15 working days. The average time will be 5 working days following receipt. Results are transmitted to users by hard-copy. However, provisional results from specimens with suspected CPE isolates are generally phoned and or emailed directly to the relevant consultant microbiologist if contact details such as mobile phone or email address are provided. [email notification is in a form that protects patient identity].

Where time and resources allow, we endeavour to keep to the times stated, however occasions may arise where there are unforeseen delays.

WGS typing results are generally available some time after the initial report because the nature of the typing method means that it is efficient to process isolates in batches. The value of this process is to establish links between cases in the same institution and across institution. At present turnaround time is approximately 2 to 3 months but there are plans to improve this through 2017.

5.0 Specimen Rejection Policy

Specimens sent to the CPERL will be rejected if:

- Unlabelled slope and/or request form
- Mislabelled slope and/or request form
- Broken slopes
- Mixed cultures

6.0 Transportation of Specimens

- Inoculated slopes should be packaged and transported according to ADR (Carriage of Dangerous Goods by Road) regulations.
- Specimens can be referred to CPERL via a courier company e.g. Hays DX, Biomnis, Capital Freight or Nightline couriers.
- A number of slopes may be sent in each crush-proof container but each slope must be individually wrapped in an adsorbent material to prevent breakage during transit.

7.0 Retention Times

- All slopes referred are stored for a minimum of 48 hours after analysis.
- All referred isolates are stored on PROTECT beads at -80°C for a minimum of 5 years.
- All DNA extracts are stored at -20°C for a minimum of 1 year.

8.0 Complaints

Consumer Affairs and the National Advocacy Unit, Quality and Patient Safety Directorate have responsibility for developing and implementing best practice models of customer care within the HSE and promotes service user involvement across the organisation through the concept of "Your Service Your Say".

Note: If users have a complaint or feel that any part of the service is unsatisfactory or could be improved on in any way they should contact the laboratory.

A complaints form is available to download:

http://www.nuigalway.ie/research/salmonella_lab/downloads/lmd_complaints_form.pdf

9.0 APPENDIX

Target genes covered by Real – Time PCR

9.1 Class A Carbapenemases

- **KPC (**KPC-1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17,18, 19)
- **GES (**GES-2, 4, 5, 6, 11, 13, 14, 15, 16, 17, 18, 20, 22, 24)
- **IMI** (IMI-1, 2, 3, 4)

9.2 Class B Carbapenemases

- **IMP** (IMP-1, 2, 5, 6, 8, 9, 10, 11, 15, 19, 20, 21, 24, 25, 29, 34, 37, 40, 41, 42, 45)
- **NDM** (NDM-1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 12, 13, 14, 15, 16, 17, 18)
- **VIM** (VIM-1, 2, 3, 4, 5, 6, 8, 9, 10, 11, 12, 14, 15, 16, 17, 18, 19, 20, 23, 24, 25, 26, 27, 28, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40)

9.3 Class D Carbapenemases

- **OXA-48** (OXA-48, 162, 163, 181, 204, 232, 244, 245, 370)
- **OXA-23** (OXA-23, 27, 49, 73, 146, 165, 166, 167, 168, 169, 170, 171, 225, 239)
- **OXA-24** (OXA-24, 25, 26, 33, 40, 72, 139, 207)
- **OXA-58** (OXA-58, 96, 164)
- **OXA-51** (OXA-51, 64, 65, 66, 68, 69, 71, 82, 90, 92, 107, 113, 128, 172, 200, 201, 202, 219)