Molecular Detection of Emerging Carbapenemases from Rectal Swab Colonization Screenings in the Northeast.

K. Cummings| S. Morris, L. Rose, L. Leach, J. Bodnar, C. Jacobsen, K. Wahl, C. Prussing, K. Musser, M. C. Rowlinson, E. Nazarian

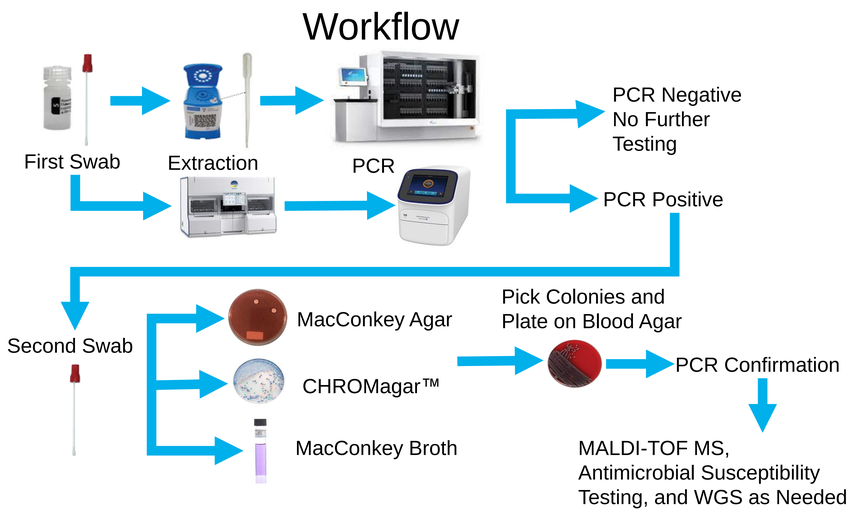
NEW YORK STATE | Department of Health | Wadsworth Center

Introduction

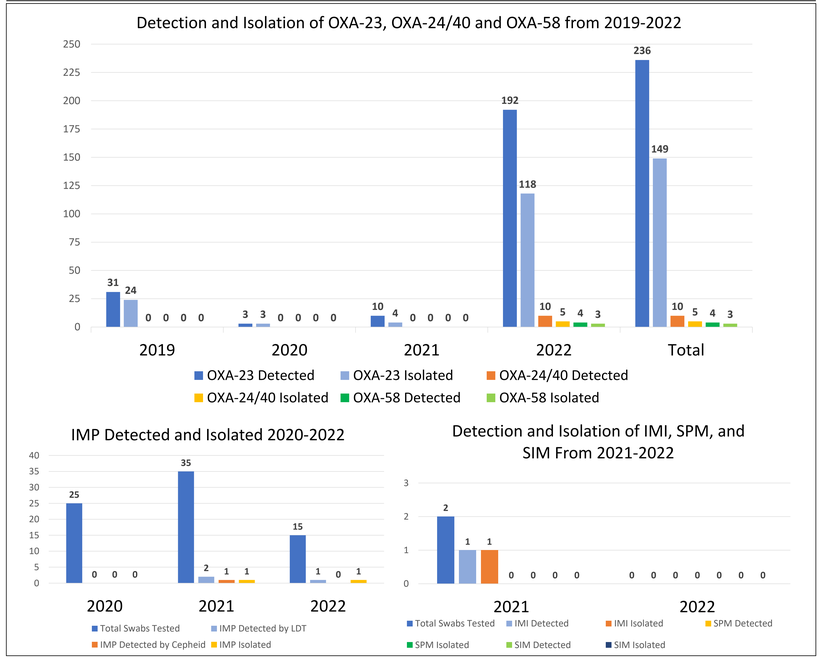
- The detection of carbapenemase-producing organisms (CPOs) impacts rapid containment and mitigates transmission in healthcare facilities (HCF). The Wadsworth Center (WC) performs testing for CPO colonization screenings (CS) throughout the Northeast (NE) United States as part of the Antimicrobial Resistance (AR) Lab Network to detect the “Big Five” carbapenemases (KPC, NDM, OXA-48, VIM, and IMP) from rectal swabs using the Cepheid Xpert® Carba-R (Carba-R) assay.
- Carba-R cannot detect all IMP variants, or other emerging carbapenemases (OXA-23, OXA-24/40, OXA-58, SIM, SPM, and IMI).
- CS to identify emerging carbapenemases currently requires culture which is labor intensive and increases time to identification.
- WC validated three multiplex real-time PCR assays to be performed on CPOs as a rapid method for identification of novel carbapenemases from rectal swabs: “OXA” (OXA-23, OXA-24/40, OXA-58), “IMP” (IMP), and “Novel” (SIM, SPM, and IMI).
- PCR-positive swabs are cultured to recover CPO for further characterization.
- Samples have been collected from the NE from 2019 to present.

Materials and Methods

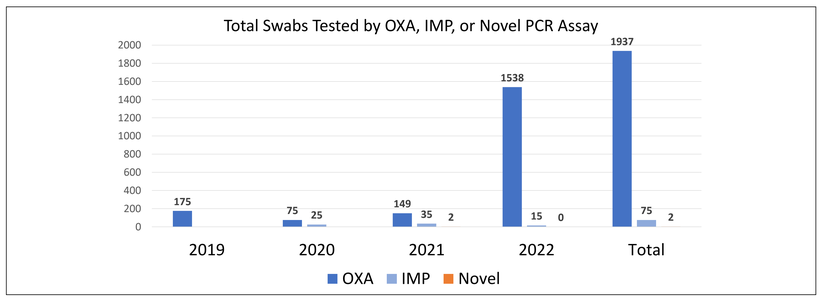
- Dual rectal swabs are collected for CS (Cepheid dual swab collection kit (catalog #900-0370).
- One swab is tested with the Carba-R assay and if positive, the remaining swab is used for culture.
- From the remaining sample in the Carba-R lysis vial inoculated from the first swab, 600 µl of the lysis reagent is used for DNA extraction with the addition of an internal control. The sample is extracted on bioMérieux’s EMAG®. (There is sufficient volume from the lysis vial to perform 2 Carba-R tests and 2 extractions if a repeat is necessary).
- Extracted DNA is tested with the appropriate PCR assay (OXA, IMP, or Novel).
- PCR-positive swabs are inoculated to CHROMagar™ mSuperCARBA™, Direct MacConkey Agar, and MacConkey broth. Suspect colonies are isolated to blood agar and tested by PCR for gene confirmation.
- Mechanism confirmed isolates are then identified by MALDI-TOF MS and antimicrobial susceptibility testing is performed.
- Whole-genome sequencing (WGS) can be performed which provides the gene variant and multi-locus sequence type from our AR WGS pipeline, and relatedness with our Legiocluster pipeline, if appropriate.



OXA, IMP, IMI, SPM, and SIM Results

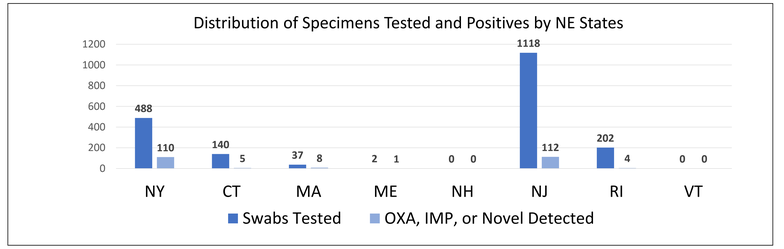


- 1937 swabs were tested by the OXA PCR assay from 2019-2022 from the Northeast Region. There were 236 OXA-23 positive swabs (12.2%), 10 OXA-24/40 positive swabs (0.5%), and 4 OXA-58 positive swabs (0.2%). From the positive swabs 149, 5, and 3 were able to be isolated for their respective mechanism.
- Seventy-five swabs were tested by the IMP assay from 2020-2022 in the Northeast Region. There were 3 IMP positive swabs. 2 of the swabs could be detected by the Cepheid Carba-R assay and were isolated by culture.
- Two swabs were tested by the Novel assay from 2021-2022. There was 1 positive IMI swab that was able to be isolated by culture.



- In 2022, the number of samples received for the OXA assay has increased 10-fold. Whereas IMP and Novel
- Note: Data from 2022 is until 9/9/22.

OXA, IMP, IMI, SPM, and SIM Results



- Specimens were received from 6 of the 8 states within the NE for the OXA, IMP and Novel CS assays.
- The majority of specimens received were from NJ and NY.

NDM/OXA-58 Outbreak Investigation

- An outbreak of NDM was identified in a NE Region facility.
- CS’s performed at the WC detected NDM using the Carba-R from a patient rectal swab specimen.
- Further WGS analysis using WC’s in-house AR WGS pipeline identified OXA-58 in addition to NDM from the *Acinetobacter baumannii* isolated recovered from the positive rectal swab.
- To date, we have detected 4 NDM/OXA-58 positive rectal swabs with 3 isolated by culture.
- Subsequent WGS analysis for relatedness determined the 3 *Acinetobacter baumannii*, NDM/OXA-58 isolates were related differing by only 1-7 mutation events.

	Patient 1	Patient 2	Patient 3
Patient 1	0	6	7
Patient 2	6	0	1
Patient 3	7	1	0

Conclusion

- The use of the OXA, IMP, and Novel real-time PCR assays for detection of emerging carbapenemase genes from rectal swabs has enhanced our CPO CS capacity by only performing culture on PCR positive samples.
- The assays can detect patients with lower CPO burdens than by culture alone.
- Our workflow of screening the swabs by PCR decreases turn-around-time to result and streamlines culture isolation and additional testing to provide data to prioritize infection prevention measures.

Acknowledgements

- We thank the Wadsworth Center Applied Genomic Technologies Core (ACTC) for whole genome sequencing and Wolfgang Haas, PhD for Bioinformatics analysis.
- This work was supported by the New York State Department of Health, Cooperative Agreement Number NU50CK000423 funded by the Centers for Disease Control and Prevention (CDC), and Cooperative Agreement U60OE000103 funded by CDC through the Association of Public Health Laboratories.