

Enhancement of Carbapenemase Producing Organism Colonization Screening Workflow: Decreasing Time, Increasing Data, and Maximizing Resource Capabilities

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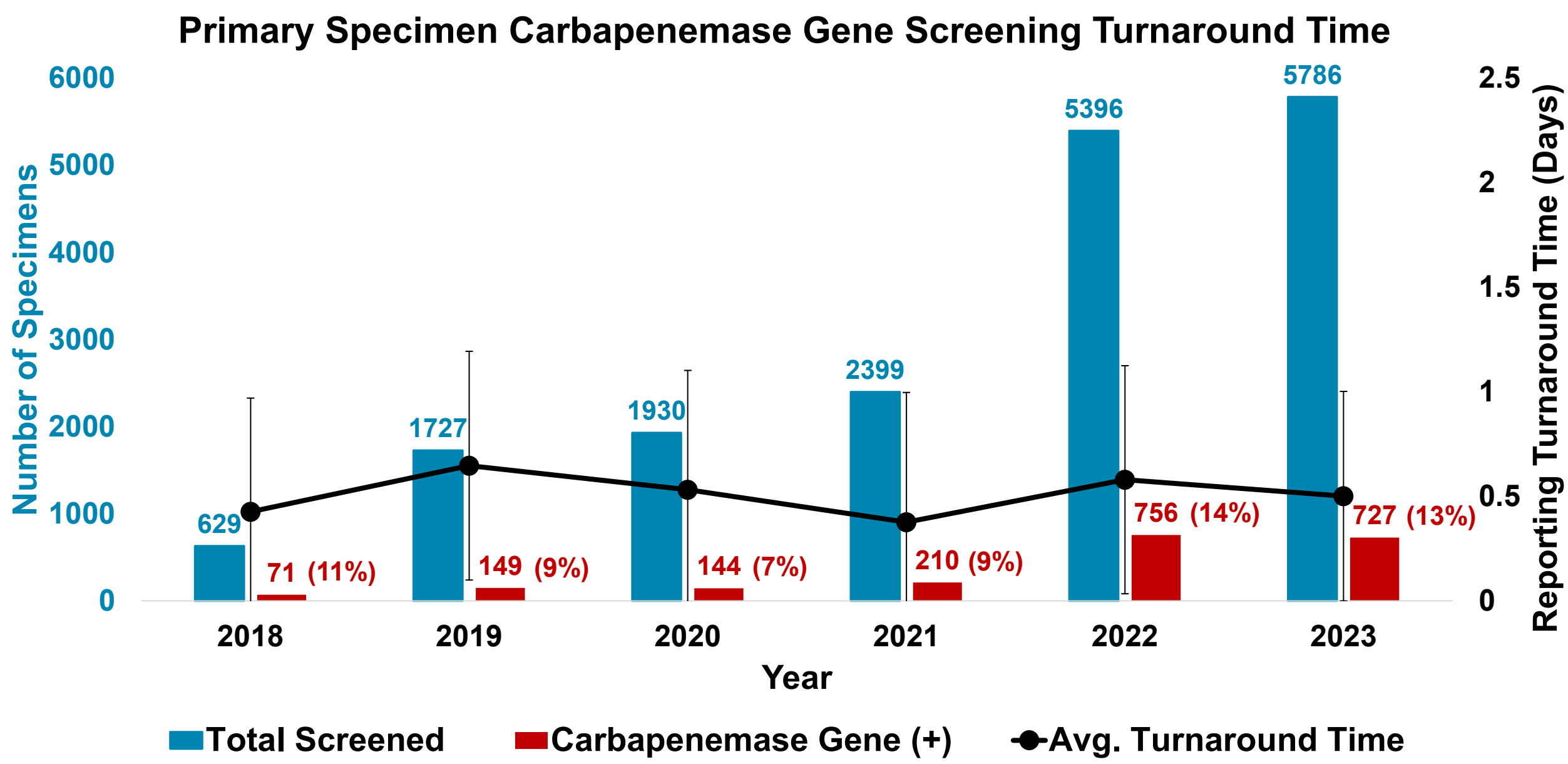
Introduction

- Carbapenems are β -lactam antibiotics, last line defenses against gram-negative bacterial infection
- The spread of carbapenem resistance, by carbapenemase producing organisms (CPOs) is a significant global health problem
- Carbapenem-resistant *Klebsiella pneumoniae*, *Acinetobacter baumannii* and *Pseudomonas aeruginosa* are among the most urgent public health threats, causing 4.5K deaths and 54K hospitalizations per year in the U.S.¹
- Detection of CPOs guides isolation and transmission mitigation procedures in healthcare facilities
- Common CPO colonization screening (CS) techniques typically begin with culturing for isolation from rectal swab specimens reflecting > 3 workdays reporting turnaround time (TAT)
- Per day, 1 unidentified colonization of carbapenem-resistant *K. pneumoniae* is predicted to transmit to 2 uncolonized individuals²
- Methods for more rapid TAT can provide greater public health impact
- The Wadsworth Center (WC) performs CPO CS throughout the Northeast as part of the Centers for Disease Control and Prevention's Antimicrobial Resistance Laboratory Network
- The WC performs CPO CS from rectal swabs for the “Big Five” carbapenemase genes (*bla*_{KPC}, *bla*_{NDM}, *bla*_{OXA-48}, *bla*_{VIM}, and *bla*_{IMP}) and carbapenemase genes associated with carbapenem-resistant *Acinetobacter* spp. (*bla*_{OXA-23}, *bla*_{OXA-24/40}, and *bla*_{OXA-58})
- Workflow beginning with multiplex real-time PCR (rtPCR) allows the WC to cost-effectively report carbapenemase gene screening results in < 24 hours

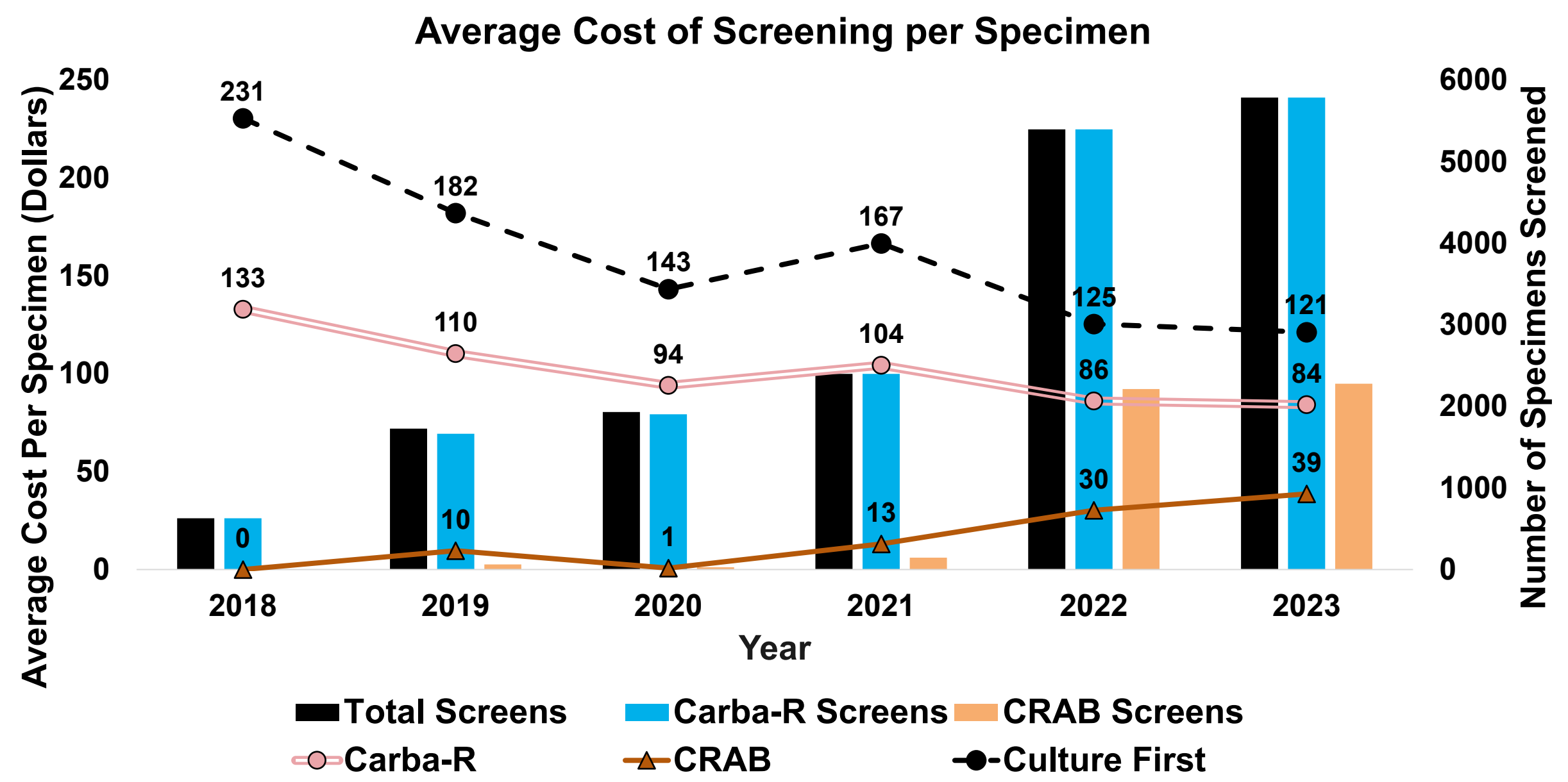
Methods

- Dual rectal swabs collected for CS (Copan Dual Swab Collection Device)
 - First swab tested using Cepheid Xpert® Carba-R (*bla*_{KPC}, *bla*_{NDM}, *bla*_{OXA-48}, *bla*_{VIM} and *bla*_{IMP} multiplex rtPCR) assay (Carba-R)
 - If alternative multiplex rtPCR is warranted*, 600 uL from Carba-R reagent using first swab is used for bioMérieux's EMAG® DNA extraction paired with multiplex rtPCR (*bla*_{OXA-23}, *bla*_{OXA-24/40} and *bla*_{OXA-58}) (CRAB)
 - If a multiplex rtPCR assay is positive, second swab is cultured using CHROMagar™ mSuperCARBA™, MacConkey Agar, and MacConkey broth, incubated at 35°C overnight
 - Suspected CPOs isolated on trypticase soy agar with 5% sheep blood plates (BAP) and tested using Hardy Diagnostics' NG-Test CARBA 5 (KPC, NDM, OXA-48, VIM and IMP immunochromographic strip) (CARBA 5); BAP plates with positive CARBA 5 or suspected *Acinetobacter* spp. are incubated at 35°C overnight
 - Carbapenemase gene verification through multiplex rtPCR, organism identification through MADLI-TOF MS and antimicrobial susceptibility testing (AST) using Thermo Scientific™ Sensititre™ Gram Negative GN7F AST Plates
- *Cepheid Xpert® Carba-R does not screen for IMP gene variation or alternative OXA gene families
- ** CHROMagar™ *Acinetobacter*™ utilized, if *Acinetobacter* spp. colonization is suspected

Primary Specimen Carbapenemase Gene Screening Reporting

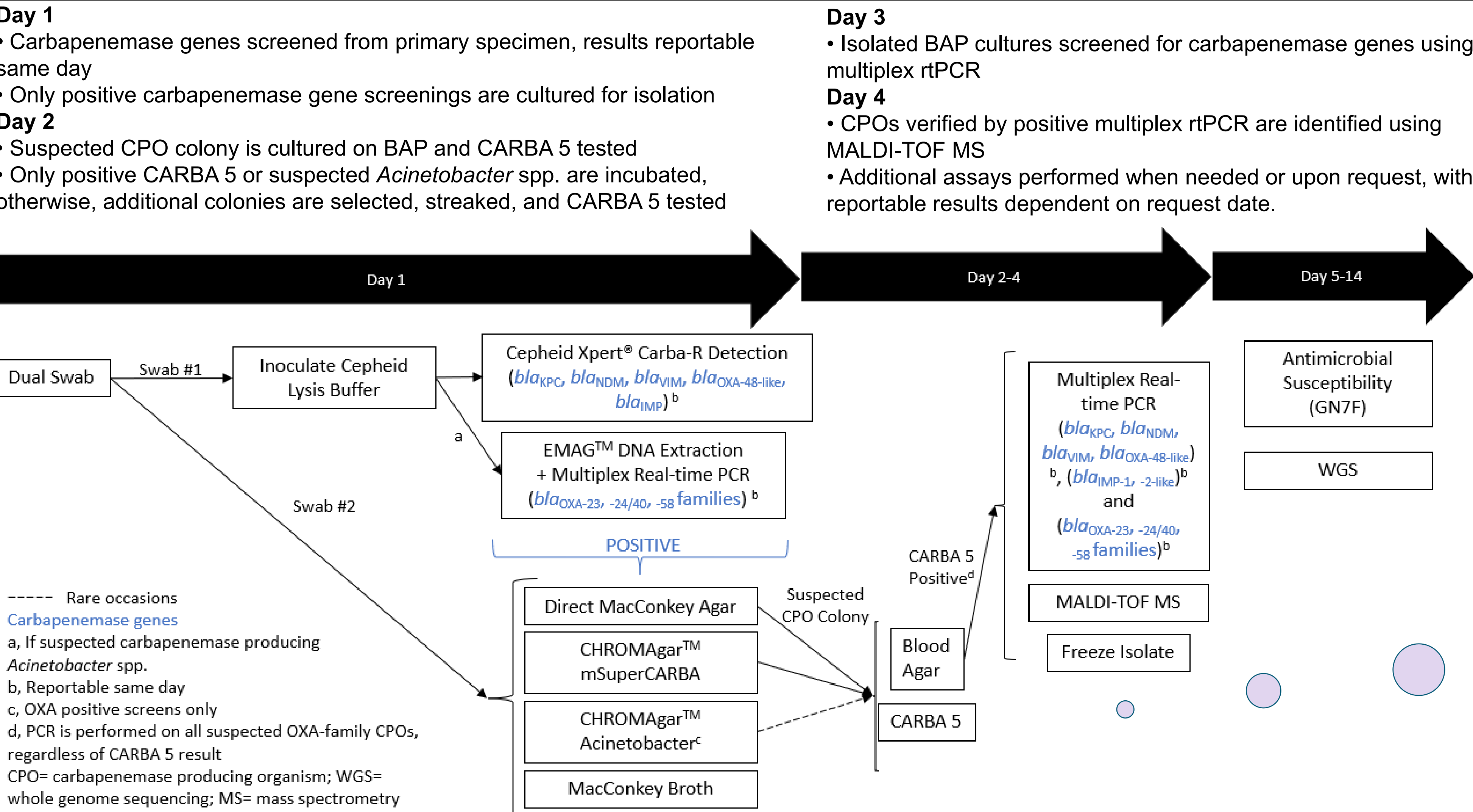


- The WC screened an increasing number of primary specimens for carbapenemase genes between 2018-2023
- The number of positive screens increased between 2018-2023
- The WC maintained TAT, averaging 0.52 days, 95% CI [0.52, 0.53], to screen primary specimens for carbapenemase genes and report results



- Materials, technician time and CPO CS reported by WC 2018-2023 were used to compare cost per specimen by Carba-R, CRAB and estimated culture-first method
- Carba-R and/or CRAB assays are consistently cost-effective initial screening methods for WC compared to a culture-first approach

Wadsworth Center CPO Colonization Screening Workflow



The WC begins CPO screening with molecular testing from the primary specimen, reporting the “Big Five” and emerging OXA family carbapenemase genes in < 24 hours

Many laboratories start CPO screening by culturing for isolation from the primary specimen taking > 3 days to report results!

Rapid Reporting Turnaround Time Contributions:

Pre-analytic	Analytic	Post-Analytic
<ul style="list-style-type: none">• Daily shipment tracking• Pre-accessioning and label printing• Labeling reagents prior to specimen receipt• AR lab technicians retrieve shipments and accession (self-accessioning)	<ul style="list-style-type: none">• Cepheid Carba-R 50 min. assay and utilizing remaining lysate in 1 – 1.5 hrs. EMAG DNA extraction for direct application in multiplex rtPCR• Auto-importing from assay systems to clinical laboratory information management system (CLIMS)	<ul style="list-style-type: none">• Electronic reporting (post supervisor review) from CLIMS to requesting agency (i.e.. Submitting healthcare facility, epidemiology, CDC, etc.)

Conclusions

- The WC’s CPO screening workflow with Carba-R and CRAB assays aids in faster carbapenemase gene screening reporting times, reduces culture workload, does not add time to traditional CPO colonization screening methods and is cost-effective.
- Additional benefits from combined assays are increased data collection and reporting, maximization of sample and reagent use, and preservation of the second rectal swab for culture testing or additional studies.
- Reporting carbapenemase gene detection from rectal swabs within 24 hours of receipt allows for rapid initiation of cohorting and transmission prevention protocols while verification of CPO colonization and identification procedures are pursued.

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References: 1) Centers for Disease Control and Prevention. Antibiotic resistance threats in the United States 2019. CDC.gov. 23 May 2024. <https://www.cdc.gov/antimicrobial-resistance/media/pdfs/2019-ar-threats-report-508.pdf> ;2) Sypsa V, Psichogiou M, Bouzala G-A, Hadjihannas L, Hatzakis A, Daikos GL. Transmission dynamics of carbapenemase-producing *Klebsiella pneumoniae* and anticipated impact of infection control strategies in a surgical unit. PLOSOne. 2012 Jul 31. doi: 10.1371/journal.pone.0041068.