

# **Microbiology of Healthcare-Associated Infections**

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2026

## Goals

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- Microorganisms causing healthcare-associated infections
- Microbiological tools that can be used to “fingerprint” microorganisms

# Properties of Microorganisms

	Size	Growth on Non-Living Media	DNA and RNA	Sensitivity to Antibiotics
Mycotic Agents				
Yeasts	3-15 $\mu\text{m}$	+	+	+
Molds	2-20 $\mu\text{m}$	+	+	+
Bacteria	1-5 $\mu\text{m}$	+	+	+
Mycoplasma	0.1-0.25 $\mu\text{m}$	+	+	+
Rickettsiae	0.3-0.7 $\mu\text{m}$	-	+	+
Chlamydiae	0.1-1.5 $\mu\text{m}$	-	+	+
Viruses	20-300nm	-	-	-

# Sources of Healthcare-Associated Pathogens

Weinstein RA. Am J Med 1991;91 (suppl 3B):179S

- Endogenous flora (SSI, UTI, CLABSI): 40-60%
- Exogenous: 20-40% (e.g., cross-infection via contaminated hands [staff, visitors])
- Other (environment): 20%?
  - Medical devices/inanimate objects
  - Contact with environmental surfaces (direct and indirect)

# Nosocomial Infections

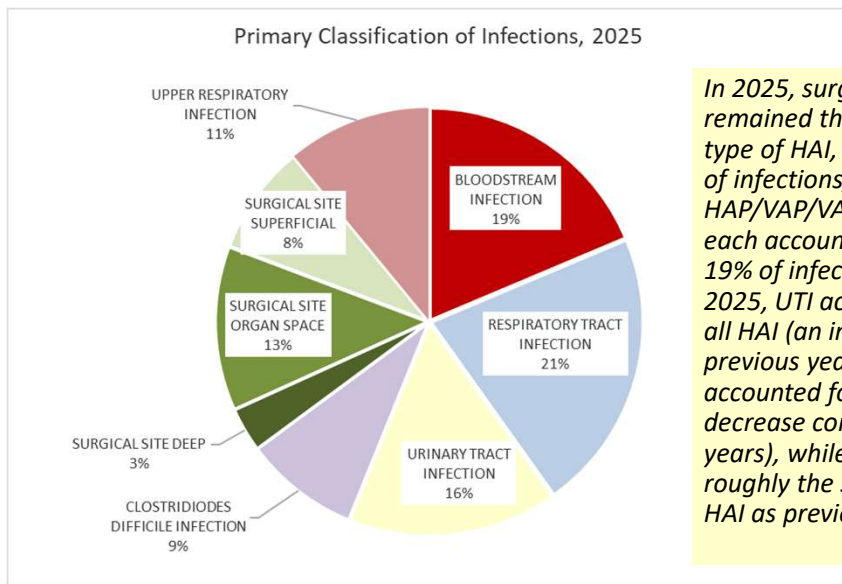
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## Chain of Infection

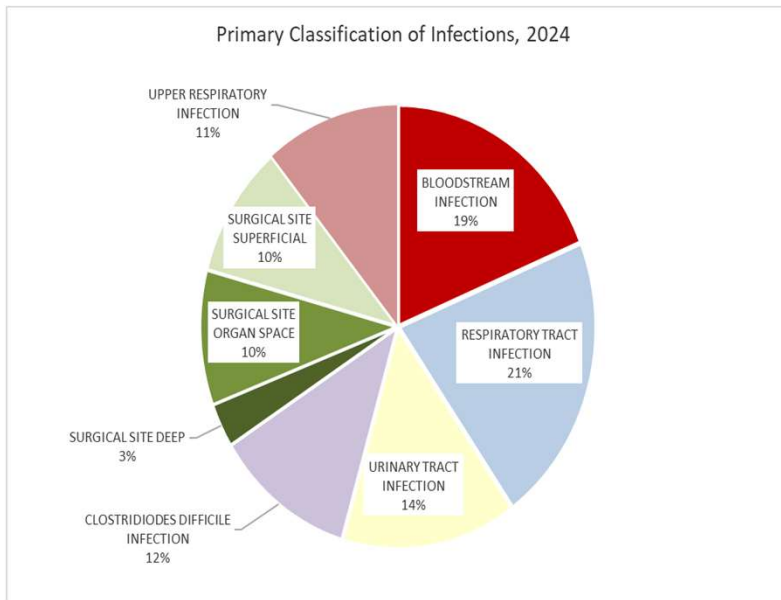
- Agent
- Mode of transmission
  - Contact (direct, indirect, droplet spread)
  - Airborne
  - Common-vehicle spread
- Host

## Mechanisms of Transmission

- Airborne-true airborne phase in route of dissemination
- Contact-victim has contact with source
  - Direct: Person-to-person (physical contact)
  - Indirect: Person-to-object-to-person (contact with contaminated intermediate object)
  - Droplet: <3 feet (brief passage of infectious agent through the air)
- Common-vehicle: Food, water, medical devices (contaminated inanimate vehicles serves as vector of agent to multiple persons)
- Vector-borne

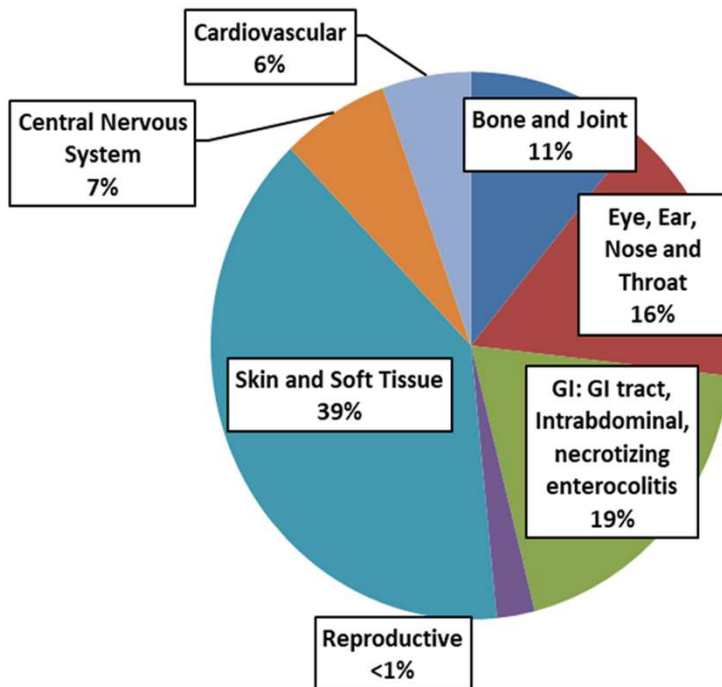


*In 2025, surgical site infections remained the most prevalent type of HAI, accounting for ~23% of infections, followed by HAP/VAP/VAE and bloodstream, each accounting for 21% and 19% of infections, respectively. In 2025, UTI accounted for 16% of all HAI (an increase from previous years) and CDI accounted for 9% of all HAI (a decrease compared to previous years), while URI accounted for roughly the same percentage of HAI as previous years*

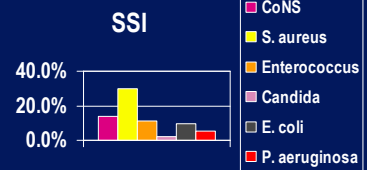
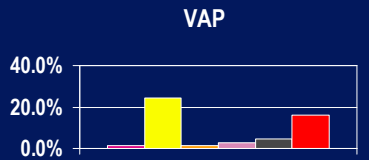
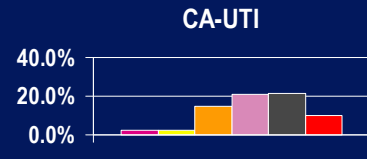
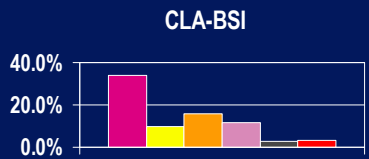


*In 2024, surgical site infections remained the most prevalent type of HAI, accounting for ~23% of infections, followed by HAP/VAP/VAE and bloodstream, each accounting for 20% and 19% of infections, respectively. In 2024, URI accounted for 11% of all HAI (an increase from previous years) and CDI accounted for 12% of all HAI (an increase compared to 2023), while UTI accounted for lower percentage of HAI than previous years.*

## Types of Infections Categorized as "Other", 2020



# PATHOGENS CAUSING HAIs, NHSN, 2006-2007



- CoNS
- S. aureus
- Enterococcus
- Candida
- E. coli
- P. aeruginosa

Hidron AI, et al. ICHE 2008;29:996-1011

# HAI PATHOGENS, NHSN, 2011-2014

Weiner LM, et al. ICHE 2016;37:1288-130

TABLE 4. Distribution and Rank Order of Pathogens Frequently Reported to the National Healthcare Safety Network (NHSN), by Type of Healthcare-Associated Infection (HAI), 2011–2014

Pathogen	Overall		CLABSI		CAUTI		VAP <sup>a</sup>		SSI	
	No. (%) of pathogens	Rank <sup>b</sup>	No. (%) of pathogens	Rank <sup>b</sup>	No. (%) of pathogens	Rank <sup>b</sup>	No. (%) of pathogens	Rank <sup>b</sup>	No. (%) of pathogens	Rank <sup>b</sup>
<i>Escherichia coli</i>	62,904 (15.4)	1	5,193 (5.4)	7	36,806 (23.9)	1	476 (5.4)	6	20,429 (13.7)	2
<i>Staphylococcus aureus</i>	48,302 (11.8)	2	12,706 (13.2)	2	2,515 (1.6)	14	2,179 (24.7)	1	30,902 (20.7)	1
<i>Klebsiella (pneumoniae/oxytoca)</i>	31,498 (7.7)	3	8,062 (8.4)	4	15,471 (10.1)	4	898 (10.2)	3	7,067 (4.7)	6
Coagulase-negative staphylococci <sup>c</sup>	31,361 (7.7)	4	15,794 (16.4)	1	3,696 (2.4)	13	72 (0.8)	13	11,799 (7.9)	3
<i>Enterococcus faecalis</i> <sup>d</sup>	30,034 (7.4)	5	8,118 (8.4)	3	10,728 (7.0)	5	32 (0.4)	21	11,156 (7.5)	4
<i>Pseudomonas aeruginosa</i>	29,636 (7.3)	6	3,881 (4.0)	10	15,848 (10.3)	3	1,449 (16.5)	2	8,458 (5.7)	5
<i>Candida albicans</i> <sup>d</sup>	27,231 (6.7)	7	5,761 (6.0)	6	17,926 (11.7)	2	193 (2.2)	10	3,351 (2.2)	12
<i>Enterobacter spp</i> <sup>c</sup>	17,235 (4.2)	8	4,204 (4.4)	9	5,689 (3.7)	9	727 (8.3)	4	6,615 (4.4)	8
<i>Enterococcus faecium</i> <sup>d</sup>	14,942 (3.7)	9	6,567 (6.8)	5	4,212 (2.7)	11	23 (0.3)	24	4,140 (2.8)	11
Other <i>Enterococcus spp.</i> <sup>d</sup>	14,694 (3.6)	10	1,974 (2.0)	14	6,291 (4.1)	7	19 (0.2)	27	6,410 (4.3)	9
<i>Proteus spp.</i> <sup>c</sup>	11,249 (2.8)	11	820 (0.8)	17	6,108 (4.0)	8	125 (1.4)	12	4,196 (2.8)	10
Yeast NOS <sup>e</sup>	10,811 (2.6)	12	763 (0.8)	18	9,443 (6.1)	6	54 (0.6)	16	551 (0.4)	25
Other <i>Candida spp.</i> <sup>d</sup>	10,641 (2.6)	13	4,730 (4.9)	8	5,178 (3.4)	10	37 (0.4)	19	696 (0.5)	19
<i>Candida glabrata</i> <sup>d</sup>	8,121 (2.0)	14	3,314 (3.4)	11	4,121 (2.7)	12	12 (0.1)	33	674 (0.5)	20
<i>Bacteroides spp.</i>	7,560 (1.9)	15	515 (0.5)	19	2 (<0.1)	30	2 (<0.1)	72	7,041 (4.7)	7
Other pathogen	51,932 (12.7)		14,130 (14.6)		9,771 (6.4)		2,507 (28.5)		25,524 (17.1)	
Total	408,151 (100)		96,532 (100)		153,805 (100)		8,805 (100)		149,009 (100)	

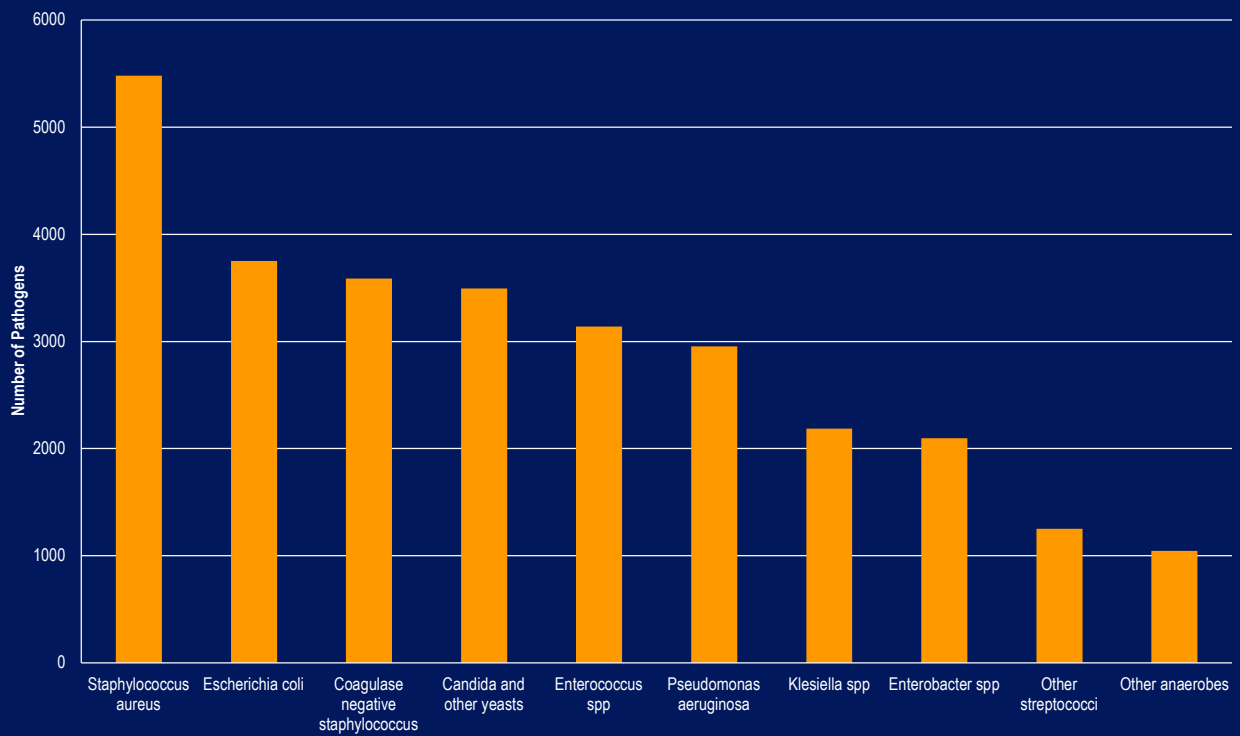
# Relative Frequency of HA Pathogens, 1980-2008

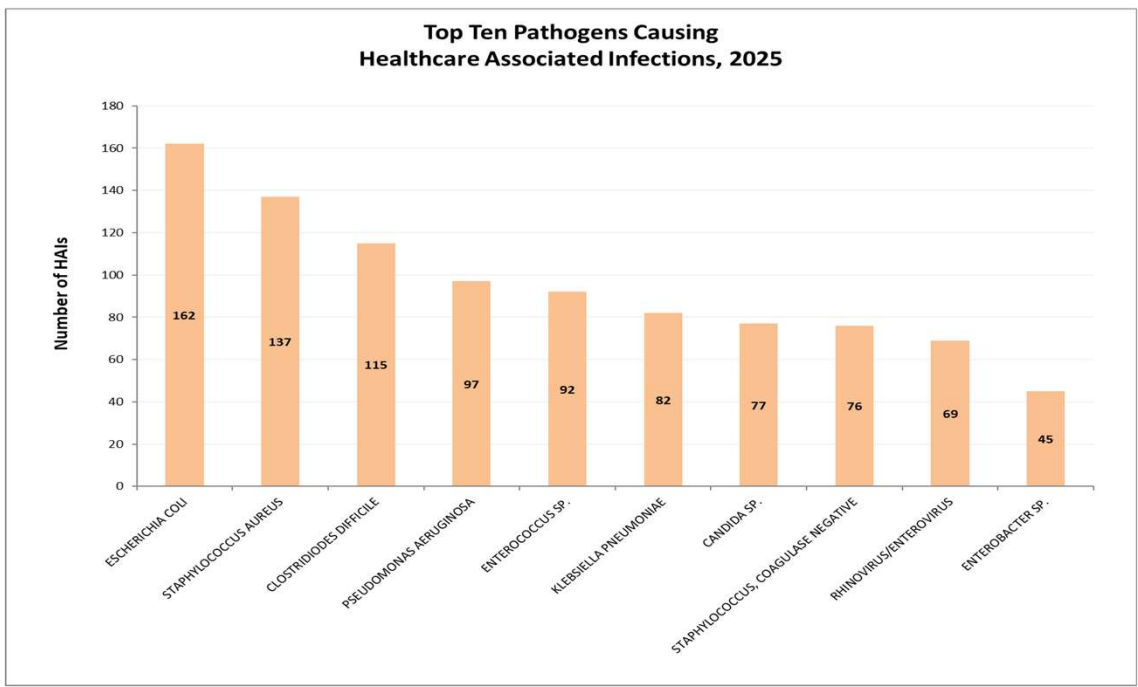
Kang, Sickbert-Bennett, Brown, Weber, Rutala. AJIC, 2012

**Table 1**  
Changes in relative frequency of health care-associated pathogens by time blocks from 1980 to 2008

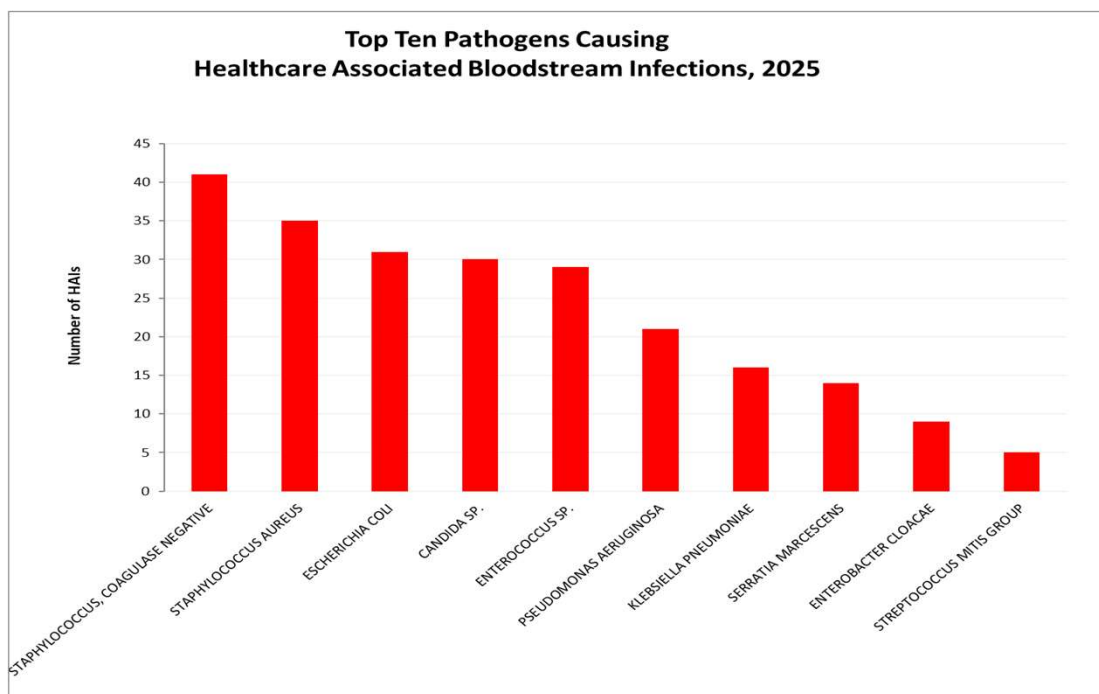
Organism	Total (1980-2008)			Percent of each time blocks						Trend analysis	
	Rank	No.	%	1980-1984	1985-1989	1990-1994	1995-1999	2000-2004	2005-2008	% Change	P value
<i>Staphylococcus aureus</i>	1	5,483	15.4	11.8	11.2	16.0	18.2	17.3	15.5	5.3	<.0001
<i>Escherichia coli</i>	2	3,753	10.6	12.6	12.7	11.3	9.2	8.2	11.5	-3.1	<.0001
Coagulase negative staphylococci	3	3,587	10.1	6.9	7.6	8.1	12.7	13.2	9.2	4.8	<.0001
<i>Candida</i> and other yeasts	4	3,494	9.8	7.7	10.4	11.0	10.3	11.1	8.1	3.0	.1890
<i>Enterococcus</i> spp	5	3,138	8.8	8.1	5.8	8.0	8.8	10.2	10.7	3.8	<.0001
<i>Pseudomonas aeruginosa</i>	6	2,954	8.3	9.5	9.5	9.7	8.6	6.7	7.1	-3.1	<.0001
<i>Klebsiella</i> spp	7	2,186	6.2	7.3	7.7	5.9	6.3	4.9	5.7	-2.4	<.0001
<i>Enterobacter</i> spp	8	2,097	5.9	7.2	8.2	6.3	4.8	4.7	5.7	-2.7	<.0001
Other streptococci	9	1,252	3.5	5.0	4.1	2.8	3.6	3.1	2.9	-1.8	<.0001
<i>Clostridium difficile</i> and other anaerobes	10	1,044	2.9	3.3	3.2	2.9	1.5	1.9	5.5	0.8	.0025
<i>Proteus</i> spp	11	946	2.7	5.4	3.9	2.1	1.6	1.9	2.1	-1.8	<.0001
<i>Serratia</i> spp	12	802	2.3	3.8	2.5	2.1	1.8	2.1	1.7	0.8	<.0001
<i>Acinetobacter</i> spp	13	593	1.7	1.2	1.4	2.2	1.4	2.1	1.6	-1.5	.0163
<i>Haemophilus</i> spp	14	494	1.4	1.6	2.5	2.2	1.1	0.9	0.8	-2.0	<.0001
<i>Bacteroides</i> spp	15	349	1.0	2.6	1.6	1.0	0.3	0.4	0.7	-0.8	<.0001
<i>Citrobacter</i> spp	16	325	0.9	1.1	1.1	0.9	0.8	0.9	0.8	0.5	.0488
Group B streptococci	17	324	0.9	1.4	1.3	1.1	0.5	0.6	0.9	-0.3	<.0001
Other	18	2,689	7.6	3.5	5.2	6.2	8.5	10.0	9.5	6.7	<.0001
Total (n)		35,510		5,217	4,336	4,904	6,964	7,999	6,090		

### Top Ten Pathogens Causing Healthcare-Associated Infections, UNC Hospitals (1980-2008)





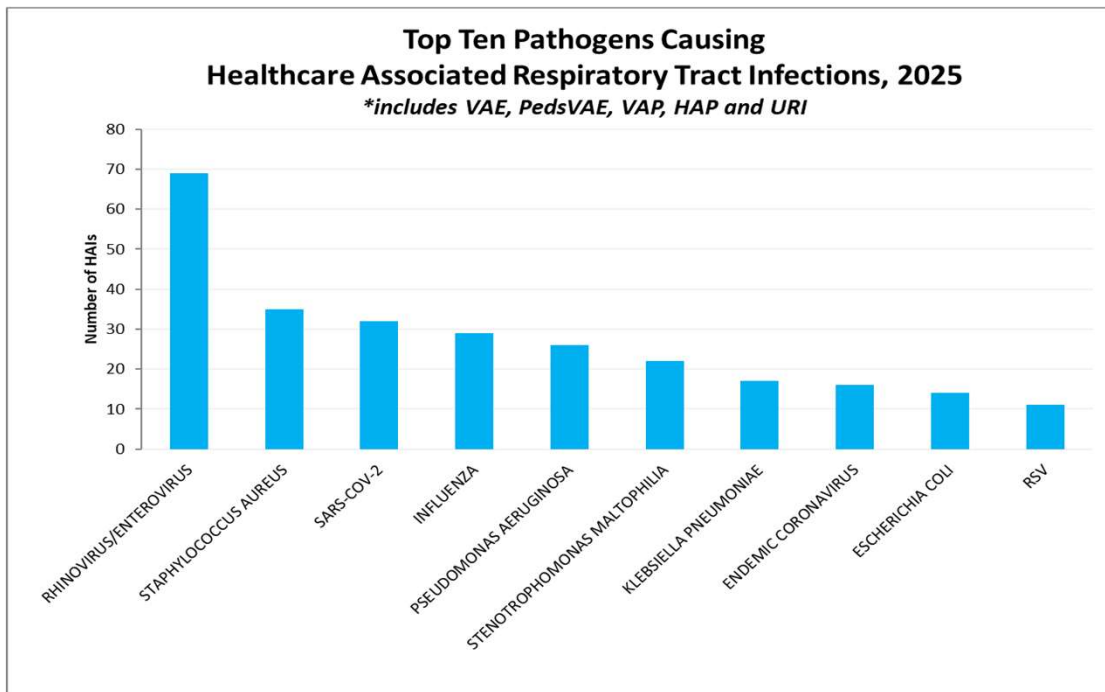
*Similar list in 2025 (same top 3 pathogens, though CDI dropped to 3<sup>rd</sup> and staph aureus moved up to 2) compared to previous recent years, however: 1) there was a ~28% increase in the number of HAI with Staphylococcus aureus, 2) there was a >30% increase in the number of Pseudomonas aeruginosa and Candida sp. HAI; 3) there was a 37% decrease in the number of SARS-CoV-2 HAI and it dropped out of top 10 list.*



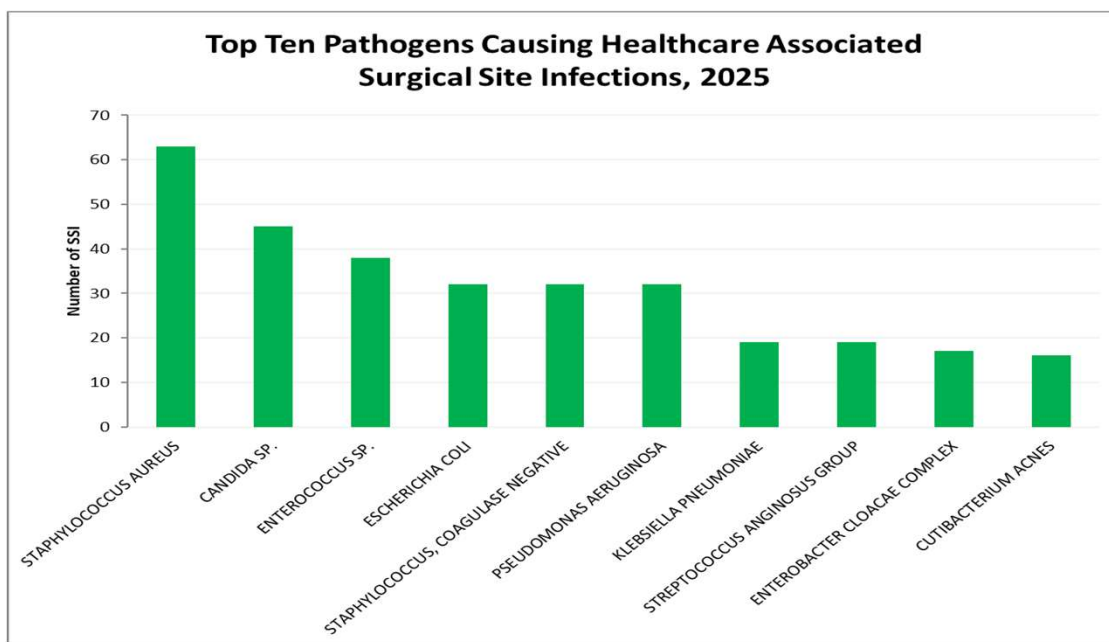
*In 2025, Staphylococcus aureus and Staphylococcus, coagulase negative accounted for a higher proportion of bloodstream infections than in 2024, while there were fewer bloodstream infections with Klebsiella pneumoniae*

### Top Ten Pathogens Causing Healthcare Associated Respiratory Tract Infections, 2025

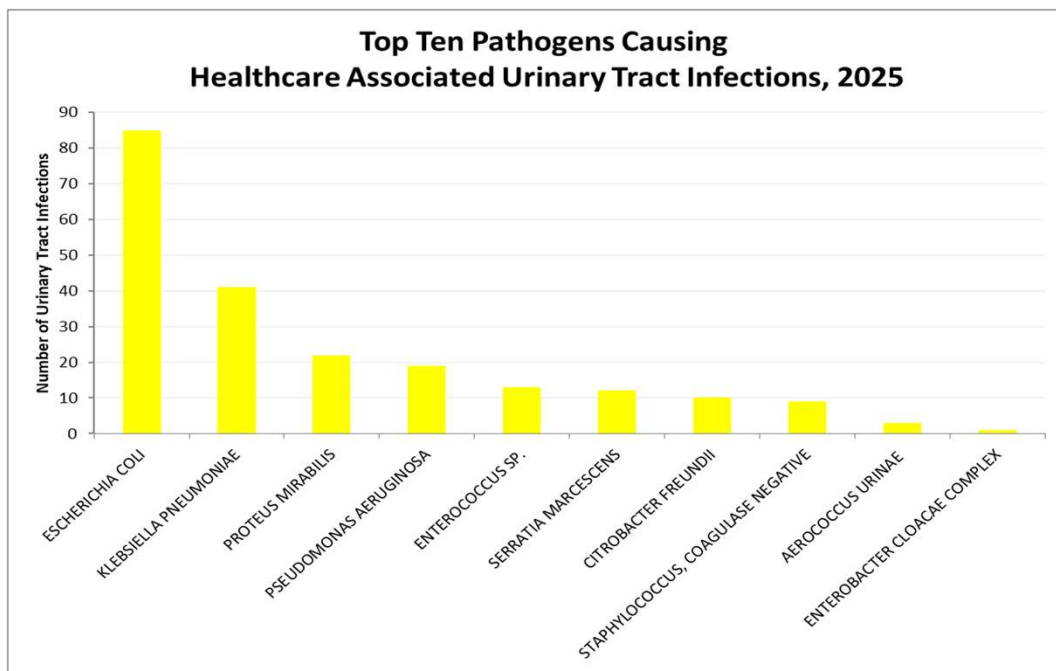
*\*includes VAE, PedsVAE, VAP, HAP and URI*



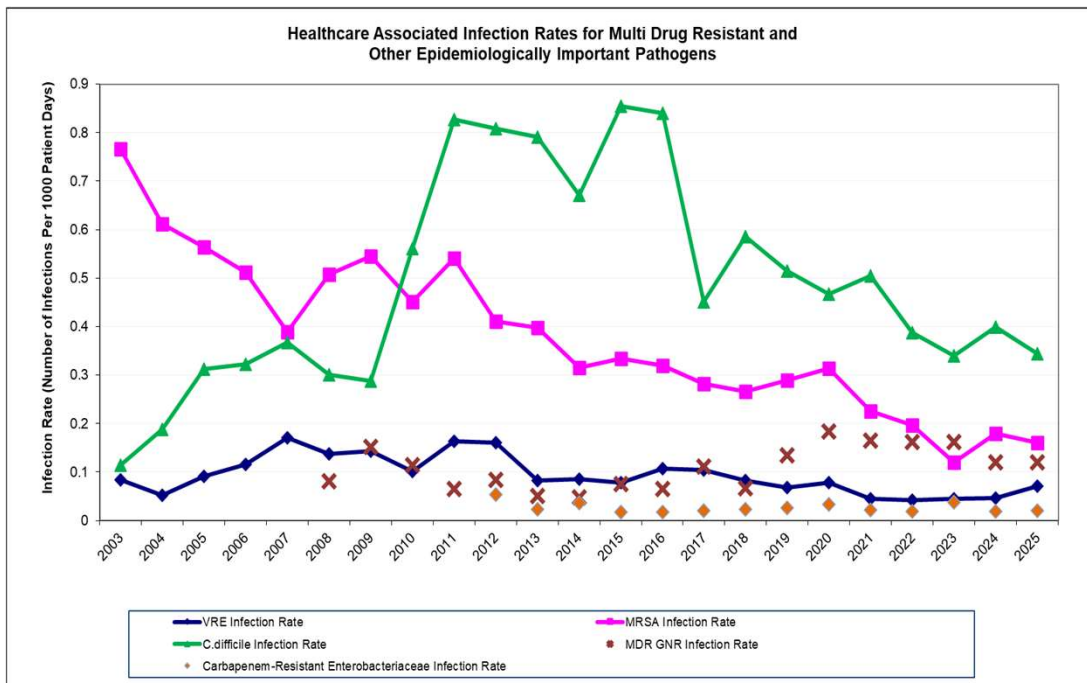
*~30% of these infections did not have a pathogen associated with it (not necessary to meet NHSN definition). In 2025, there were more respiratory HAI with rhinovirus/enterovirus, influenza and Stenotrophomonas maltophilia and fewer with SARS-CoV-2. Otherwise, proportions of HAI with remaining pathogens remained the same.*



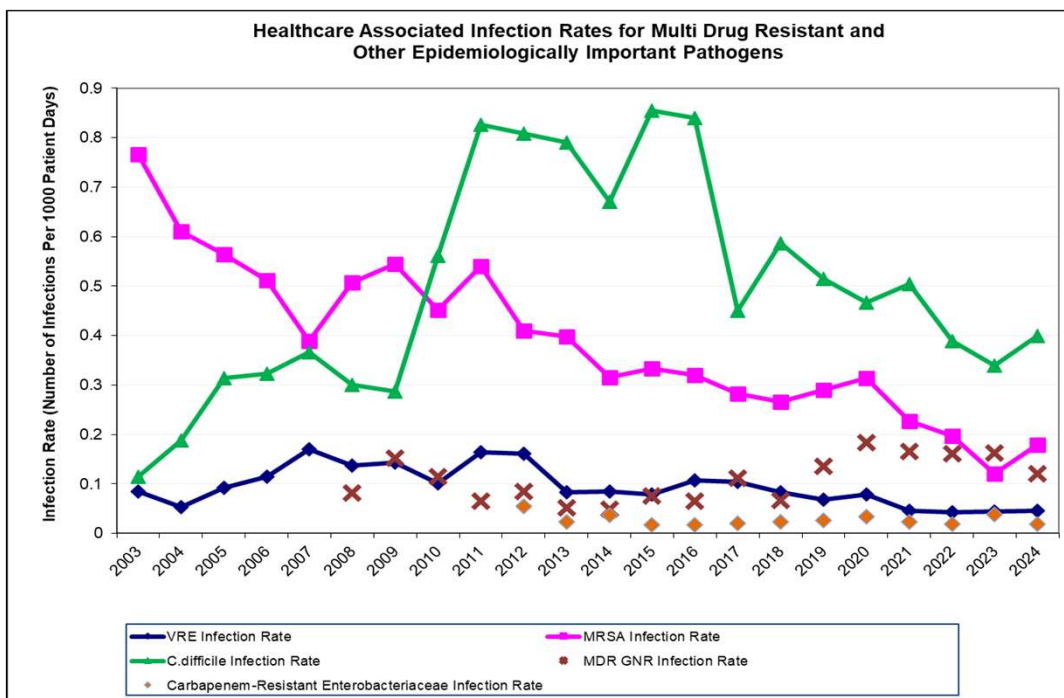
*~10% of these infections (lower percentage than previous years) did not have a pathogen associated with it (not necessary to meet NHSN definition). Of those infections with an organism, ~33% are skin flora. The number of SSI HAI with candida sp. increased in 2025. Otherwise, proportions of these pathogens did not shift in 2025 compared to 2024.*



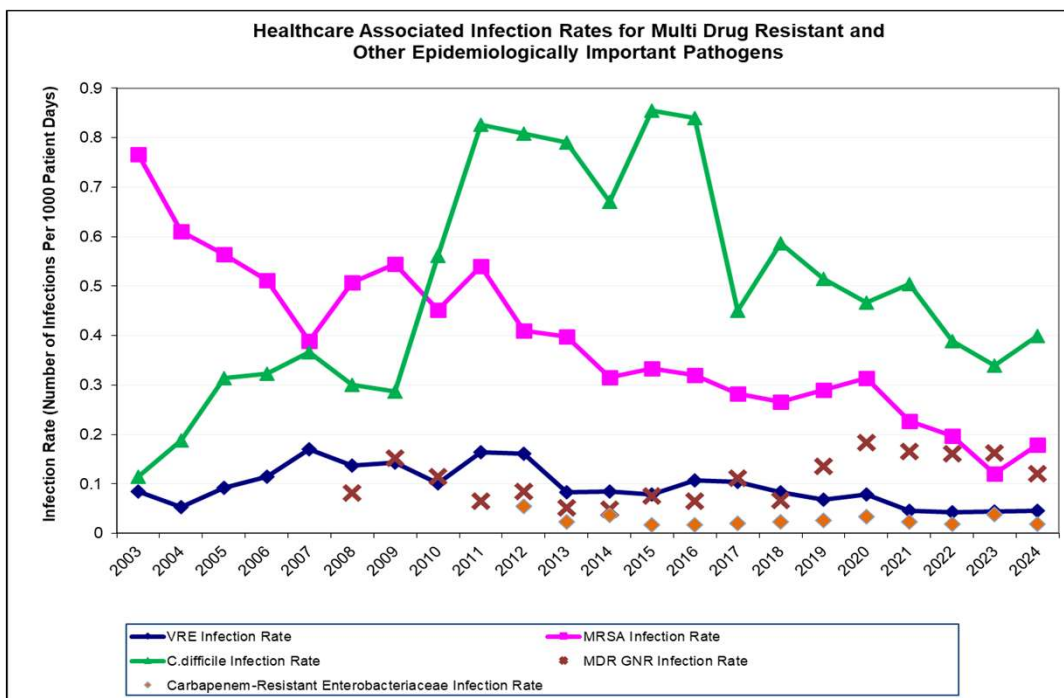
*Escherichia coli* again accounted for >40% of all 2025 hospital-associated urinary tract infections. Proportions of these pathogens



*In 2025, infection rates per 1000 patient days remained relatively stable for MDR-GNR and CRE infections while rates for MRSA and CDI infections slightly decreased and rates for VRE infections slightly increased*



*In 2024, after several years of steady decreases, the MRSA and C. difficile infection rates per 1000 patient days increased. The infection rates per 1000 patient days remained relatively stable for VRE and infections for MDR-GNR and CRE slightly decreased.*

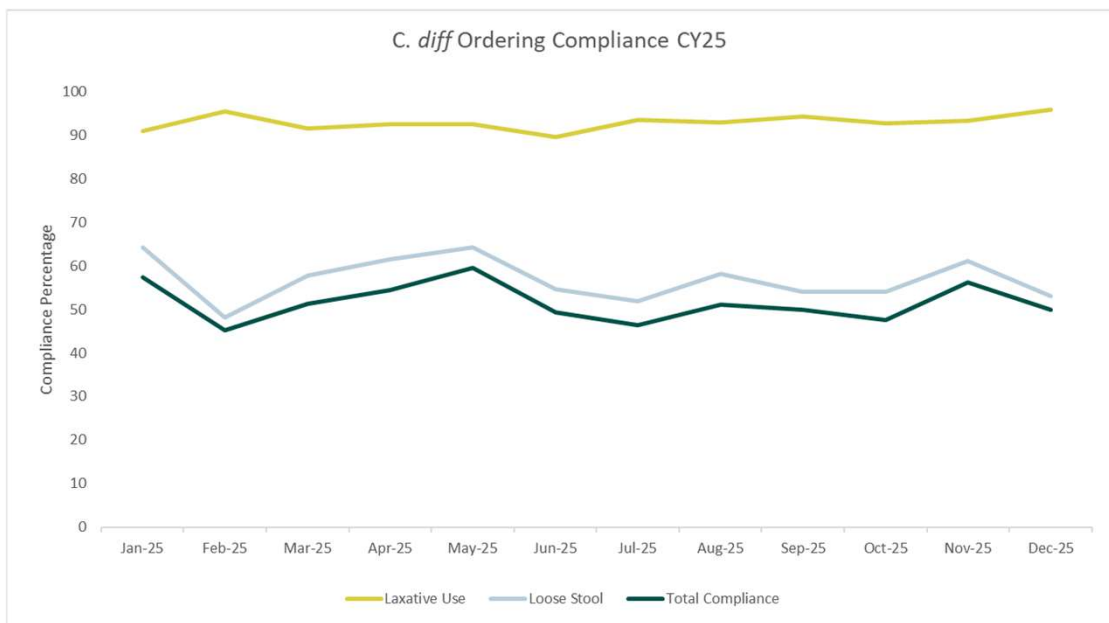


*C. difficile* increase in 2010 associated with reduced susceptibility to antibiotics, frequent antibiotic use, hypervirulent *C. difficile* strain. After experiencing a significant decrease in 2017, a slight increase in 2018, *C. difficile* HAI rates remained stable in 2021. In 2021, UNC had 33% fewer CDI infections than predicted when compared to the 2015 national baseline experience (most recent available national benchmarking data)

# Bundled Approach to Reduce *C. difficile*

(two-step GDH-glutamate dehydrogenase/toxin and NAAT-nucleic acid amplification) (2015-2017)  
Schultz et al. J Clin Microbiol 2018;56:1-10

- Diagnostic Stewardship
  - Only unformed liquid stool tested
  - No testing from patients with positive *C. difficile* test in previous 14 days
  - Testing restricted for patients with negative *C. difficile* in previous 7 days
  - Discouraged testing patients who received laxatives and/or stool softeners in previous 48 hours
- Enhanced Isolation-to 30 days after cessation of antibiotics
- Environmental C/D-standardized plan; UV
- Antimicrobial stewardship-reduce 3<sup>rd</sup>/4<sup>th</sup> generation cephalosporins and fluoroquinolones
- Hand Hygiene-clean in, clean out; immediate feedback



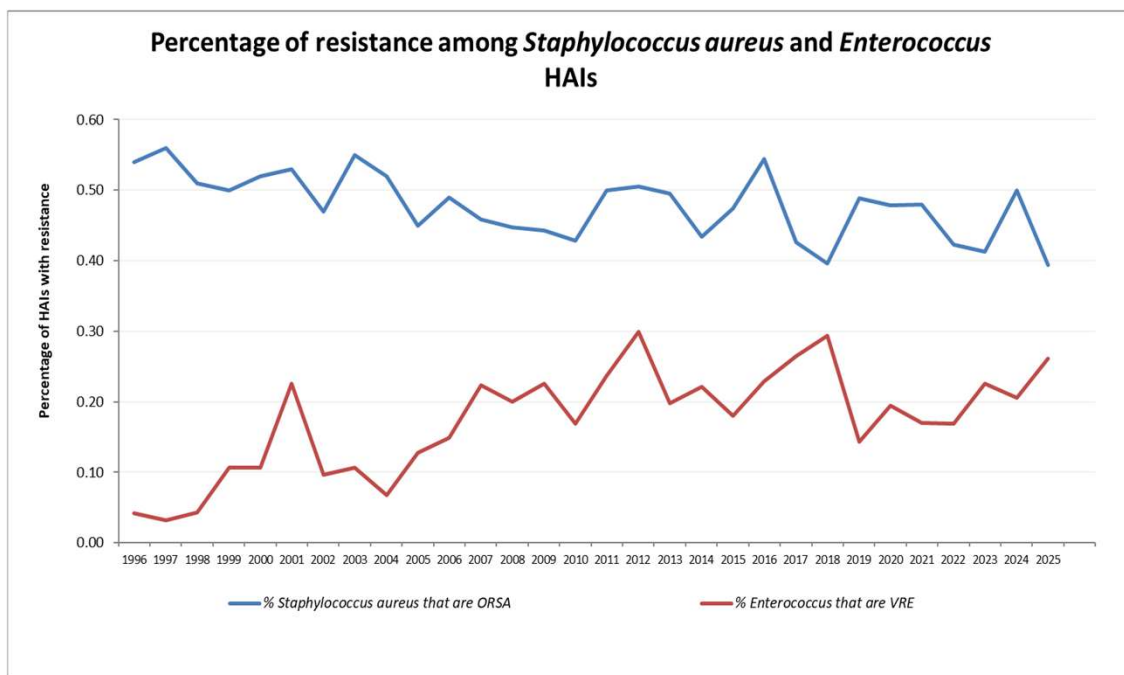
In CY25, compliance in laxative use remained >90% each month. Compliance with loose stool documentation ranged from ~48% to 64%—representing a decrease compared to CY24.

## Enteric Peer Audits CY2025

### *Percentage of Compliance*

	Jul-25	Aug-25	Sep-25	Oct-25	Nov-25	Dec-25
Enteric Isolation Sign	100	100	100	100	93	95
Bleach Wipes Available	73	98	97	92	79	90
PPE Available	91	100	100	100	88	93
PPE Compliance (Gown and gloves used)	61	74	94	92	95	84
Shared Equipment Cleaned	100	93	80	88	91	92
Soap and Water Hand Hygiene	64	91	93	97	95	88
Feedback Provided	64	95	76	88	69	84
Total Number of Audits	11	42	33	24	42	88
Unique Observers	5	11	11	6	12	20

*Enteric isolation sign consistently >90% compliance since launch of Enteric Peer Audits in July. Focus on providing feedback is needed.*



*In 2025, the percentage of resistance among Enterococcus HAIs increased in while the percentage of resistance among Staphylococcus aureus HAIs decreased compared to the past few years*

## Pathogens of Epidemiologic Importance

- **Carbapenem resistant**
  - 4 of 45 (9%) *Enterobacter cloacae*
  - 2 of 162 (1.2%) *Escherichia coli*
  - 1 of 82 (1.2%) *Klebsiella pneumoniae*
- **Multi-drug resistant**
  - 9 of 45 (20%) *Enterobacter cloacae*
  - 3 of 26 (12%) *Serratia marcescens*
  - 9 of 97 (9%) *Pseudomonas aeruginosa*
  - 11 of 162 (7%) *Escherichia coli*
  - 6 of 82 (7%) *Klebsiella pneumoniae*
  - 1 of 24 (4%) *Klebsiella aerogenes*
- **Other drug resistant**
  - 24 of 92 (26%) *Enterococcus* were vancomycin resistant
  - 54 of 137 (39%) *Staphylococcus aureus* were oxacillin resistant

*These percentages are relatively similar to percentages for CRE and MDR reported in past few years*

2025

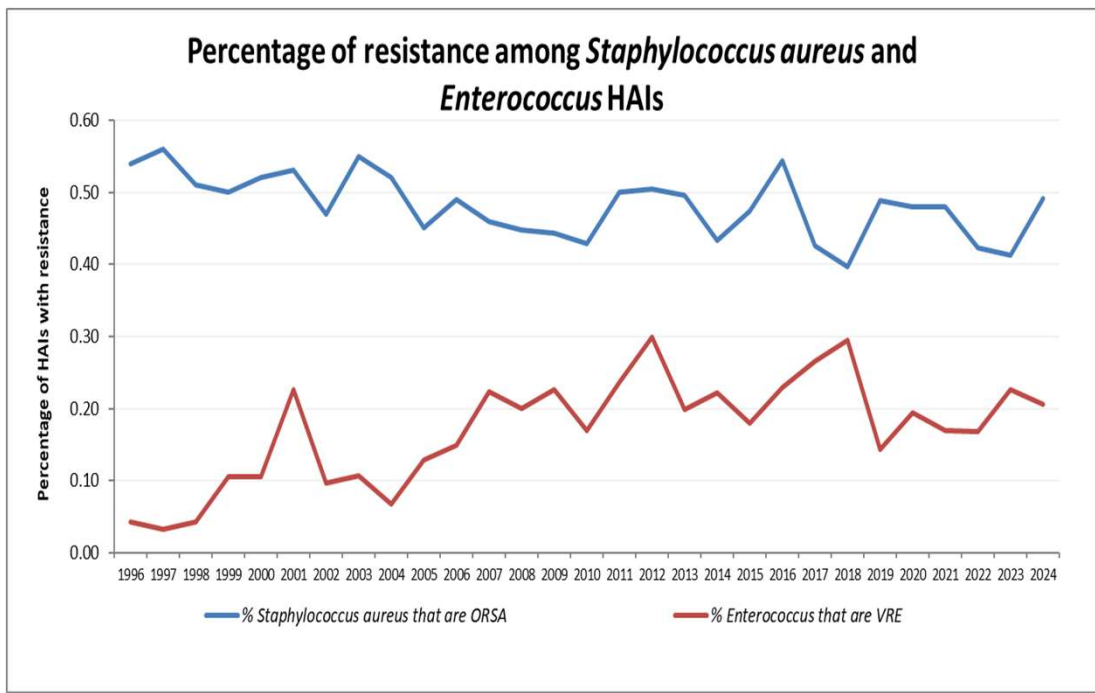
## Other Pathogens of Epidemiologic Importance

- 115 *Clostridioides difficile*
- 69 Rhinovirus/enterovirus
- 32 SARS-CoV-2
- 29 Influenza A
- 18 Parainfluenza viruses
- 16 endemic coronaviruses
- 11 Respiratory Syncytial virus
- 11 Metapneumovirus
- 10 Group B *Streptococcus*
- 9 Adenovirus
- 7 *Haemophilus influenzae*
- 7 *Mycobacterium abscessus*
- 5 *Aspergillus sp.*
- 2 *Candida auris*
- 1 *Mycobacterium fortuitum*

2025-increase in viruses likely due to viral respiratory panel (PCR), which includes multiple viruses.

## Conclusions and Recommendations

- HAI pathogens recovered at UNC Hospitals are similar in spectrum to nationally reported data.
- The proportion of vancomycin-resistance among *Enterococcus* and the VRE HAI rate increased in 2025 compared to 2024.
- The proportion of oxacillin-resistance among *Staphylococcus aureus* and the MRSA HAI rate decreased in 2025 compared to 2024.
- HAI rates of MDR Gram negative bacteria and CRE remained stable in 2025 and remain low.



*The percentage of resistance among *Enterococcus* HAIs slightly decreased in 2024 (when compared to 2023) while the percentage of resistance among *Staphylococcus aureus* HAIs increased in 2024 compared to past couple of years.*

## Pathogens of Epidemiologic Importance

- **Carbapenem resistant**
  - 3 of 28 (11%) *Enterobacter cloacae*
  - 2 of 131 (1.5%) *Escherichia coli*
  - 1 of 80 (1.3%) *Klebsiella pneumoniae*
- **Multi-drug resistant**
  - 5 of 28 (18%) *Enterobacter cloacae*
  - 16 of 131 (12%) *Escherichia coli*
  - 1 of 9 (11%) *Acinetobacter sp.*
  - 8 of 73 (11%) *Pseudomonas aeruginosa*
  - 1 of 10 (10%) *Klebsiella aerogenes*
  - 4 of 80 (5%) *Klebsiella pneumoniae*
- **Other drug resistant**
  - 15 of 73 (20.5%) *Enterococcus* were vancomycin resistant
  - 58 of 116 (50%) *Staphylococcus aureus* were oxacillin resistant

*These percentages are relatively similar to percentages for CRE and MDR reported in past few years*

2024

## Other Pathogens of Epidemiologic Importance

- 129 *Clostridioides difficile*
- 54 Rhinovirus/enterovirus
- 51 SARS-CoV-2
- 14 Parainfluenza viruses
- 10 Respiratory Syncytial virus
- 9 endemic coronaviruses
- 9 Adenovirus
- 8 Group B *Streptococcus*
- 4 Influenza A
- 4 *Haemophilus influenzae*
- 3 *Aspergillus sp.*
- 3 Metapneumovirus
- 1 *Fusarium*
- 2 *Mycobacterium abscessus*
- 1 *Mycobacterium mucogenicum*
- 1 *Mycobacterium chelonae*

2024

# Conclusions and Recommendations

UNC Medical Center, 2024

- | HAI pathogens recovered at UNC Hospitals are similar in spectrum to nationally reported data.
- | The proportion of vancomycin-resistance among *Enterococcus* decreased slightly and the VRE HAI rate remained relatively stable in 2024 compared to 2023.
- | The proportion of oxacillin-resistance among *Staphylococcus aureus* and the MRSA HAI rate increased in 2023 compared to 2023.
- | HAI rates of MDR Gram negative bacteria and CRE decreased in 2024 and remain low.

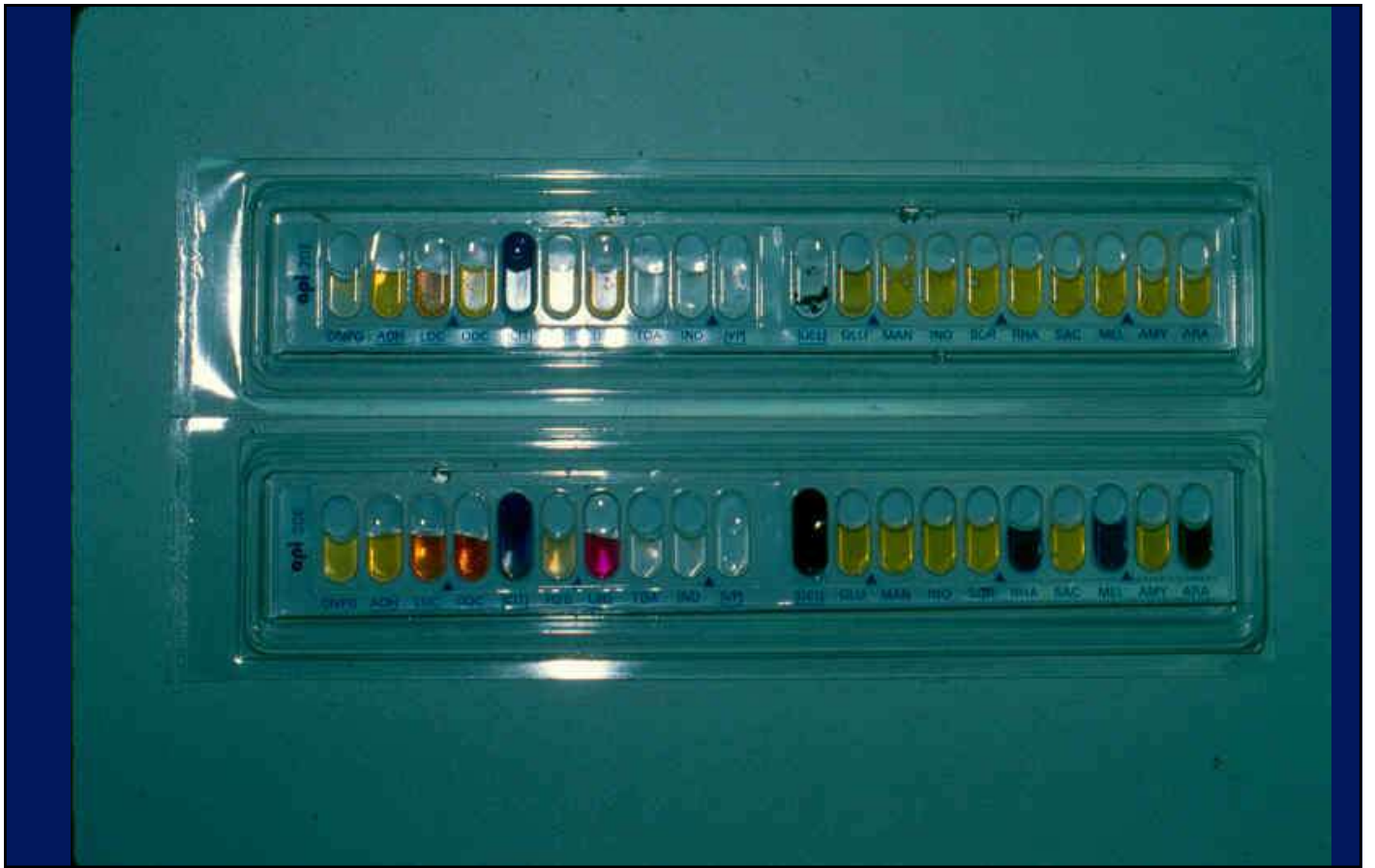
# Goals

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- Microorganisms causing healthcare-associated infections
- Microbiological tools that can be used to “fingerprint” microorganisms

## Microbiological Tools That Can Aid an Infection Preventionists

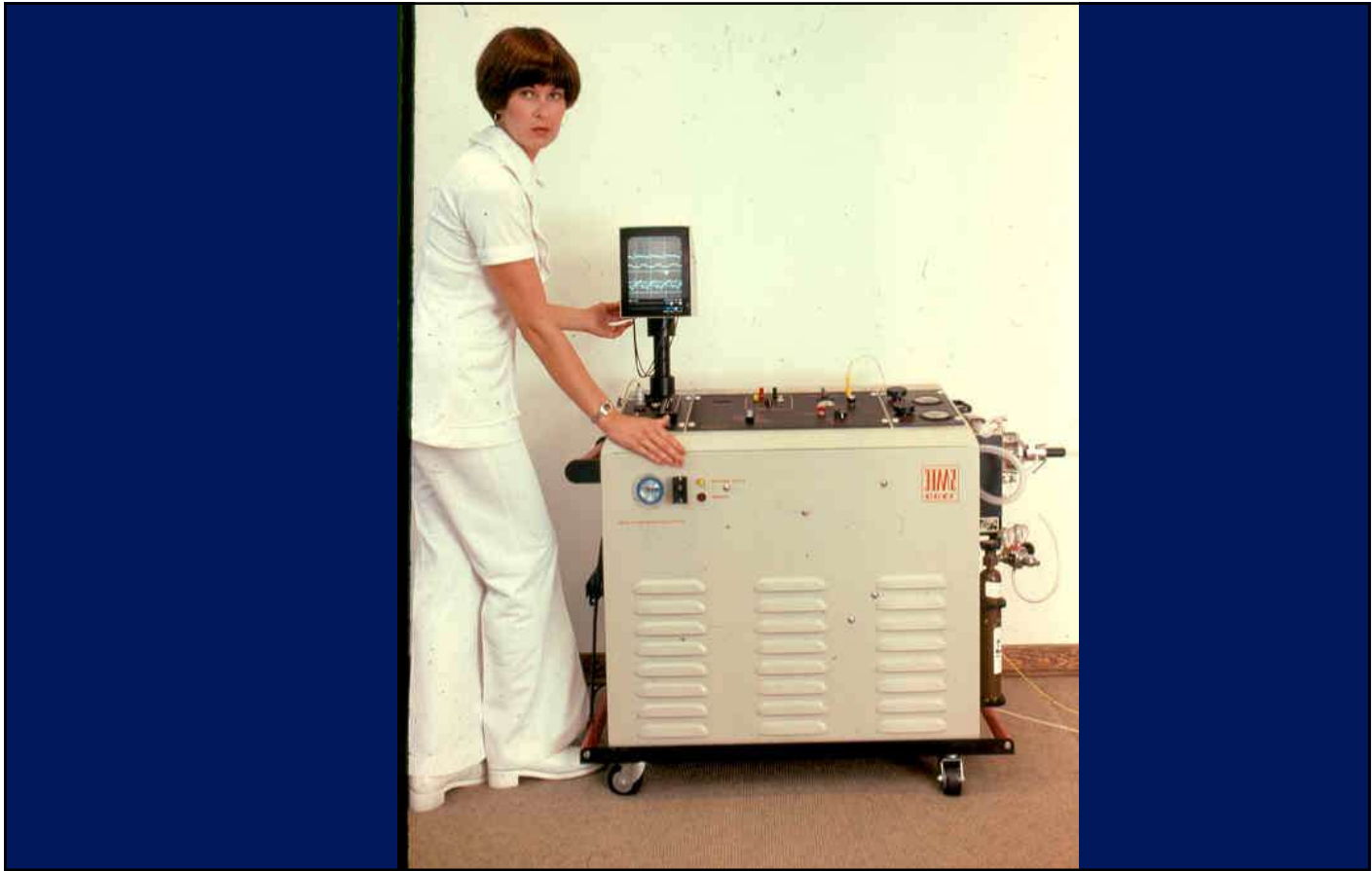
- Biotyping: use of biochemical reactions to differentiate bacteria
- Antibiograms: antimicrobial susceptibility
- Phage Typing: certain bacteria under bacterial phage attachment and subsequent lysis
- Serotyping: whole microorganisms or its components can be used as antigenic sources for a variety of serologic schemes
- Molecular Typing: microbial DNA fingerprinting (e.g., PFGE, whole genome sequence analysis-gold standard)
- MALDI-TOF mass spectrometry for organism ID (new tools bring efficiencies to ID process; peptide map used to search sequence database)







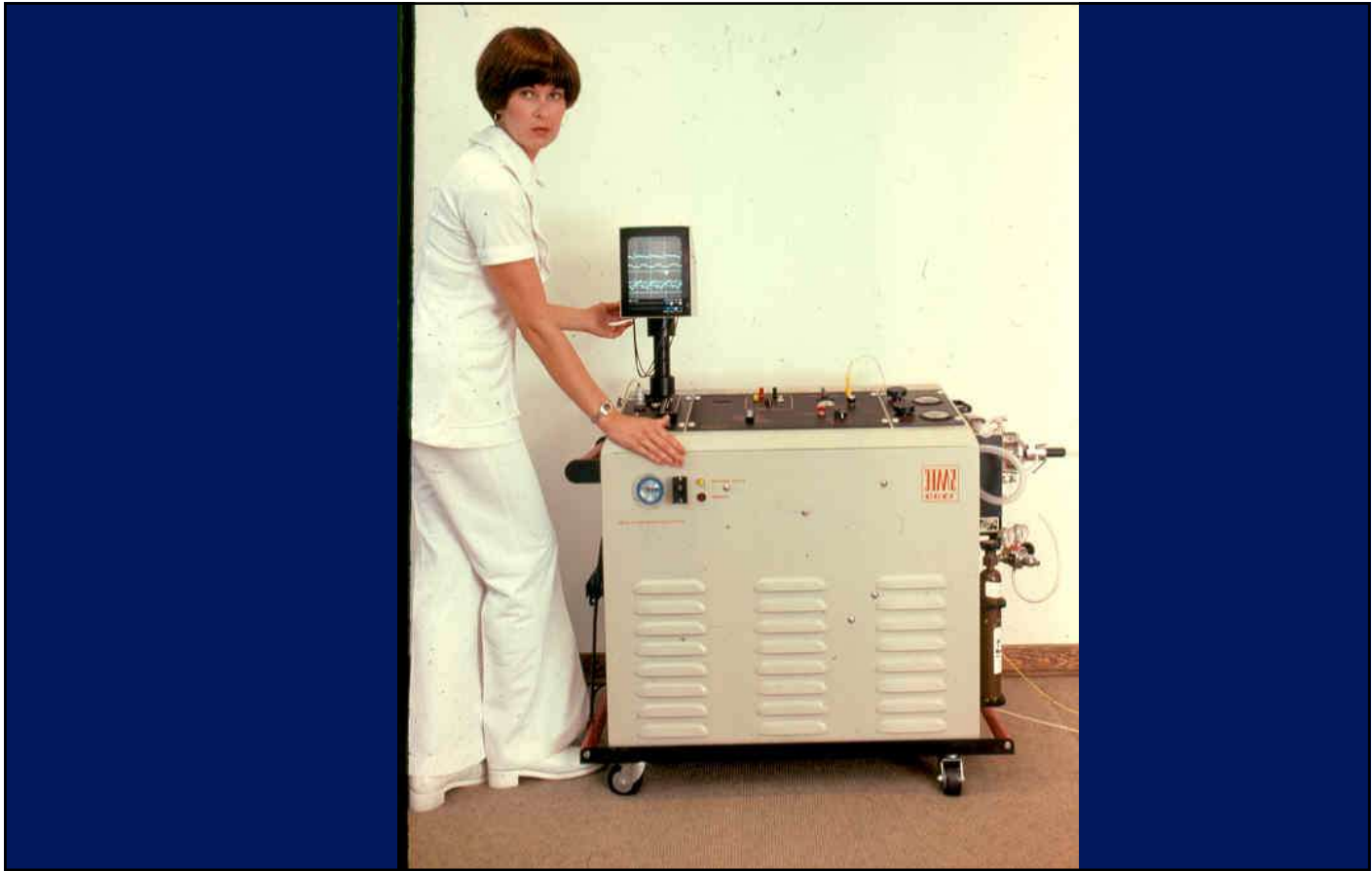




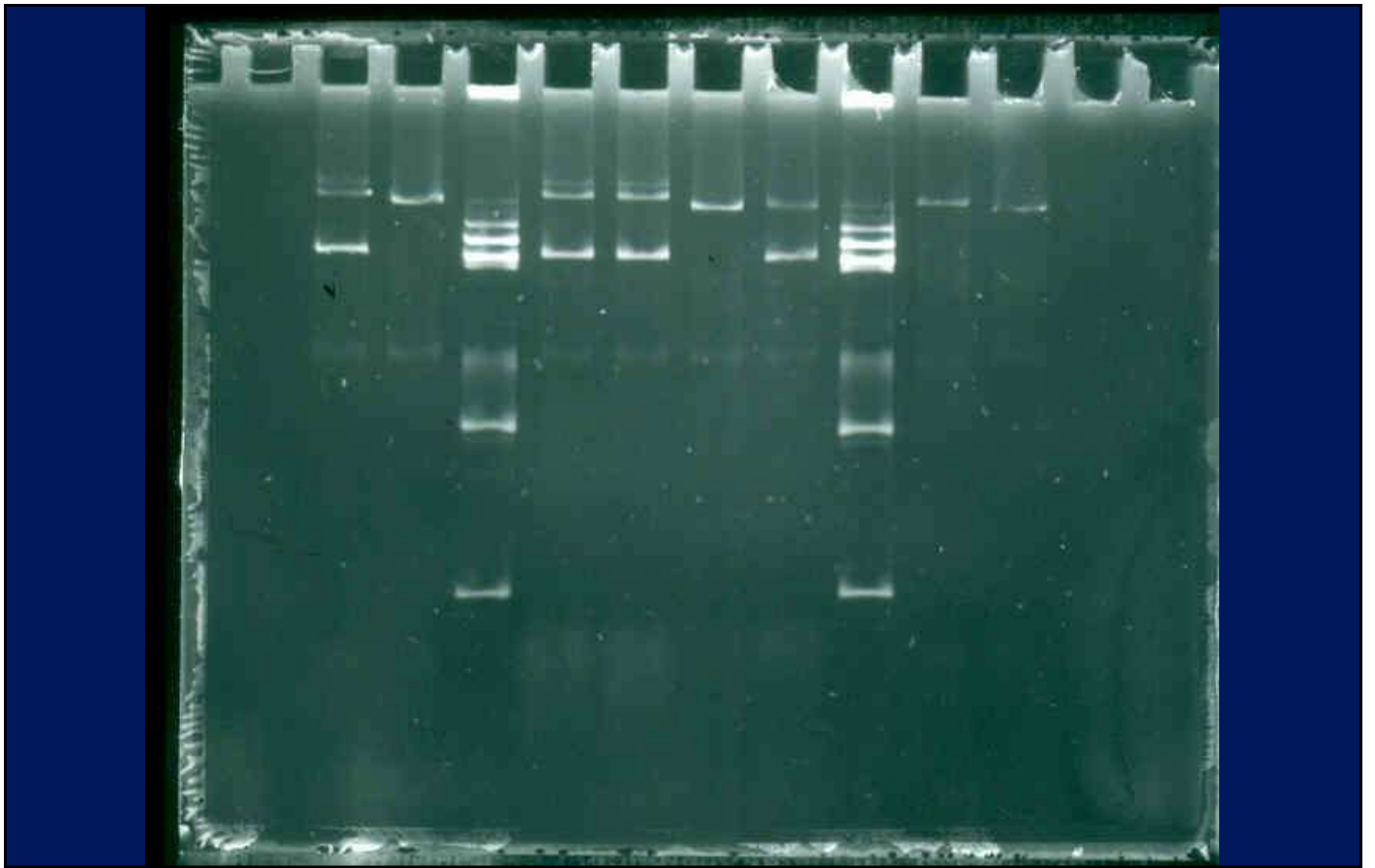
## Outbreak of *P. cepacia* Bacteremia Associated with IABP

Rutala et al. J Thoracic Cardio Surg 1988

- Cluster: Symptomatic *P. cepacia* (*Burkholderia cepacia*) bacteremia in 2 patients in CTICU within 3 days after insertion of IABP
- Evaluation: Both patients needed IABP for circulatory support
- Results: IABP water reservoir contained more than  $10^5$ /ml *P. cepacia*. Also recovered from purge button, on-off switch, hands of HCP who manipulated the water reservoir
- Agarose gel electrophoresis of *P. cepacia* revealed 3 identical plasmids
- Transmission from workers hands to patients occurred by inoculation of intravascular lines during management







# Faucet Aerators: Source of Patient Colonization with *S. maltophilia*

Weber, Rutala et al. AJIC

- Cluster of patients in SICU C/I with *S. maltophilia*
- Environmental isolates obtained from water sources
- Two isolates of *S. maltophilia* were identical to strains isolated from the faucet aerators in sinks in the patients' rooms (lanes 3,4 and lanes 6,7-patient and sink aerator)
- Believed low-level contamination of water led to contamination of faucet aerators with amplification on the aerator.
- If clusters of C/I linked to aerators, consider given to routine disinfection or removal



# Whole Genome Sequencing

## Heater Cooler Units

### | Current manufacturers

- LivaNova (Sorin)
- Maquet
- Cardioquip
- Terumo
- Cincinnati-Sub-Zero



# SOURCE OF *M. CHIMAERA* OUTBREAK

- | Point-source contamination of 3T HCU suggested by 2 studies
  - Europe: *M. chimaera* isolates from 5 patients, 3T HCU from 3 different countries and from new 3T HCU and environment at manufacturer facility – identical by sequencing (typing unpublished – preliminary)
  - US: *M. chimaera* isolates from 11 patients and 5 3T HCU from PA and Iowa were the same by whole genome sequencing (WGS). WGS \$1M in 2007, \$1,000 in 2014, today \$200-\$600/sample.
- | Manufacturing facility added disinfection and active drying procedures to production line in Sept 2014 due to *M. chimaera* contamination

Contamination during production of heater-cooler units by *Mycobacterium chimaera* potential cause for invasive cardiovascular infections: results of an outbreak investigation in Germany, April 2015 to February 2016

*Mycobacterium chimaera* Contamination of Heater-Cooler Devices Used in Cardiac Surgery – United States

Haller S, et al. Euro Surveill 2016;21(17), April 28 Perkins KM, et al. MMWR 2016;65:1117

**THANK YOU!**  
[www.disinfectionandsterilization.org](http://www.disinfectionandsterilization.org)

