Rapid Molecular Diagnostics & Antibiotic Stewardship

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Agenda

- Introduction
- Discuss the role of clinical microbiology in diagnostic stewardship.
- Understand the impact of newer technology on diagnostic and antimicrobial stewardship.
- Describe examples of day-to-day stewardship activities that clinical microbiologists perform.
- Goals of an Effective Antimicrobial Stewardship Program
- Review of current rapid diagnostic tests: Pros and Cons

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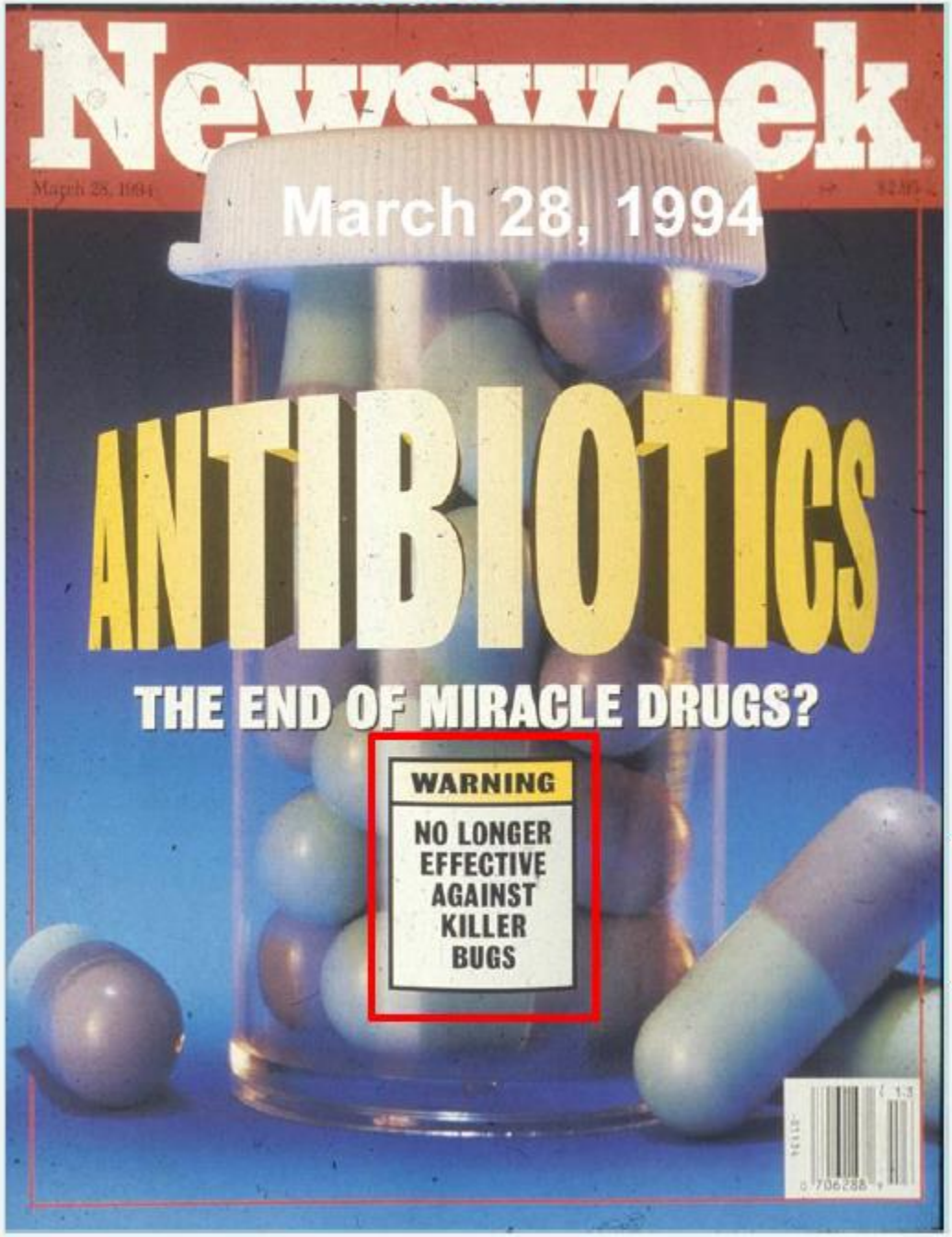
1994



2015

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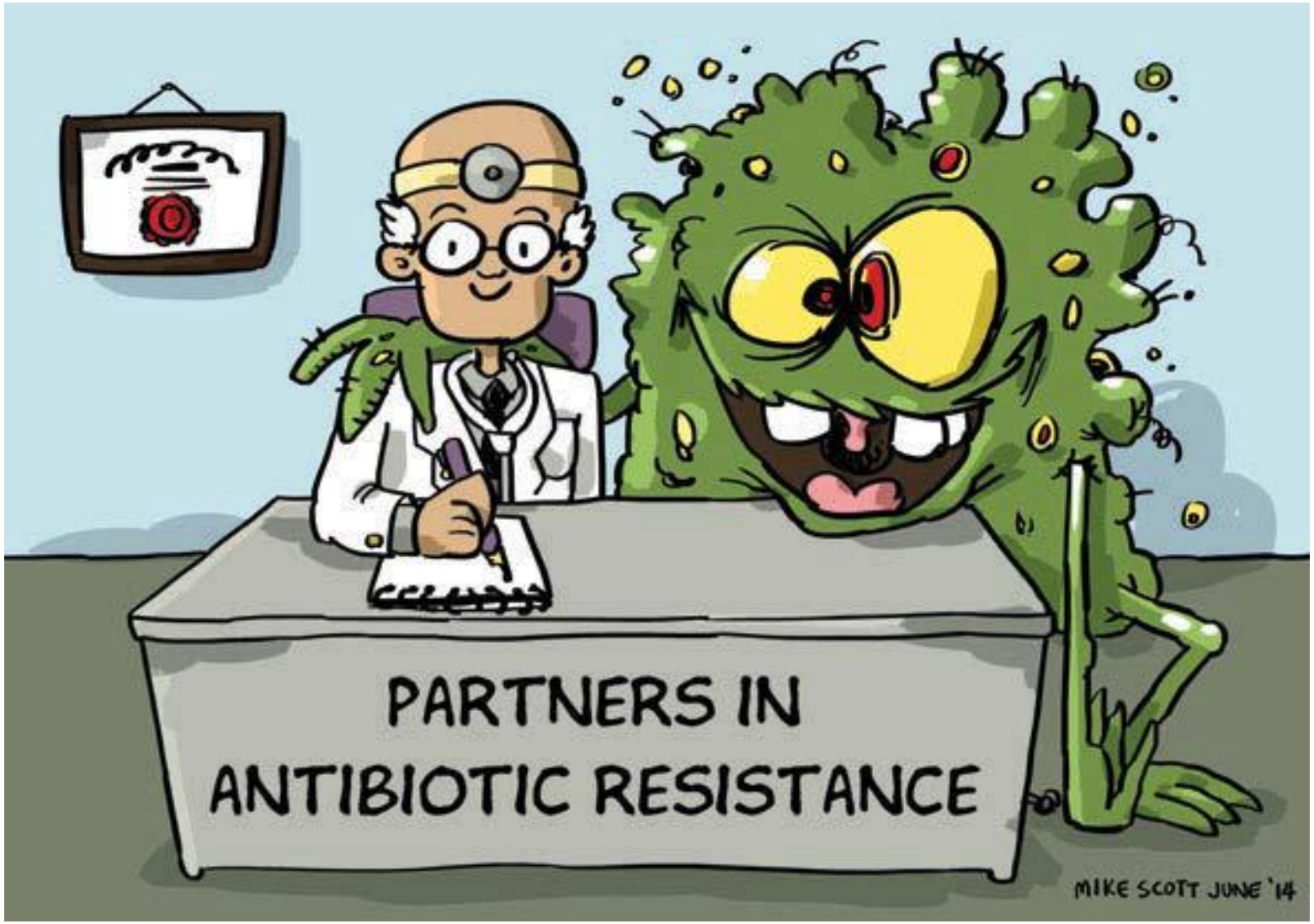
PARTNERS IN
ANTIBIOTIC RESISTANCE
MIKE SCOTT JUNE '14

Complex issue

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The New York Times

DEADLY GERMS, LOST CURES

A Mysterious Infection, Spanning the Globe in a Climate of Secrecy

The rise of Candida auris embodies a serious and growing public health threat: drug-resistant germs.

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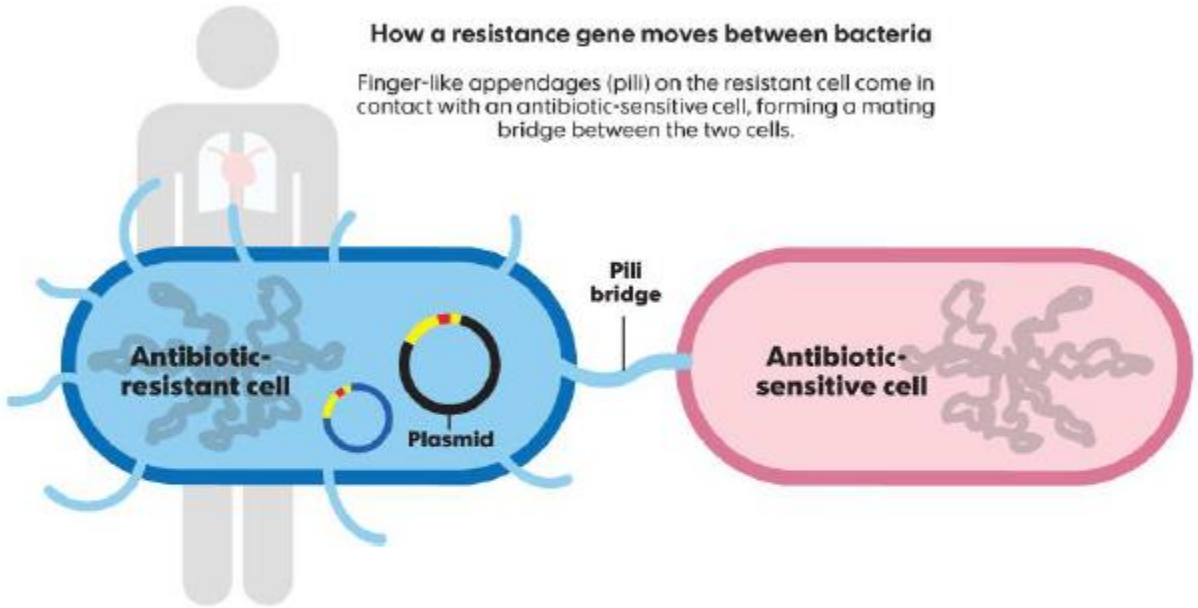
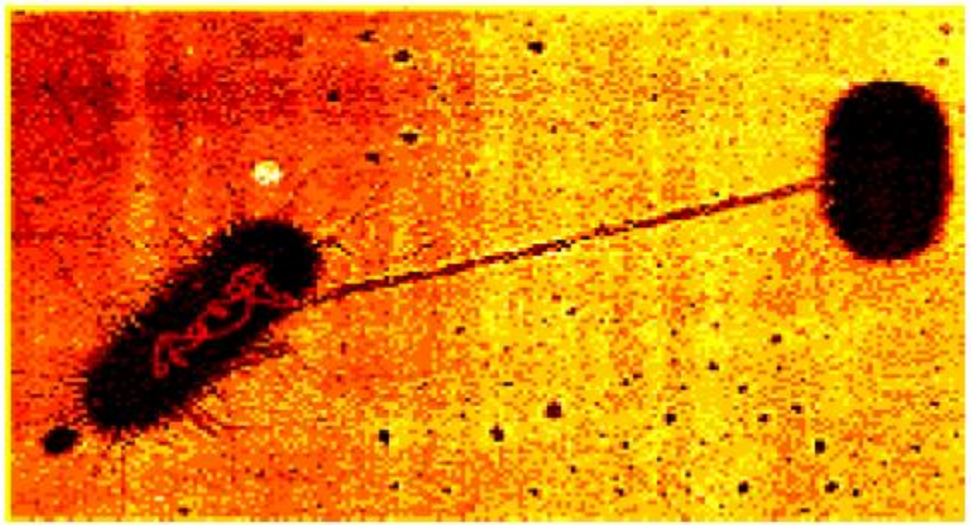
Resistance is Spreading Across Countries





- **A real global crisis**
- December 2015
- Pan-Resistant Enterobacteriaceae seen in 19 countries
- *mcr-1* >> Colistin resistant
  - Plasmid mediated
  - Easily passed between organisms (E. coli/Klebsiella)
  - Pan Resistance = No drugs work

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Why We Need to Improve Antibiotic Use

- Antibiotics are misused across the continuum of care- **up to 50% of antibiotic use is either unnecessary or inappropriate across all type of health care settings**
- Use of antibiotics **in animals-~80% of antibiotics sold in US** are **used in animals primarily to promote growth and prevent infection**
- Molecular methods have confirmed that resistant bacteria in animals are consumed by humans resulting in infection
- **Up to 90% of antibiotics used in animals are excreted in urine and stools** and **can disperse in fertilizer, groundwater, and surface runoff**
- Antibiotic misuse adversely **impacts patients and society ↑Antimicrobial resistance(AR)** and *C difficile infections*
- In 2011 a national survey found that **60% of infectious diseases physicians had seen a pan-resistant, untreatable infection in the last year 3**
- Improving antibiotic use **improves patient outcomes and saves money**
- Improving antibiotic use is a public health imperative-WHO considers AR an emerging threat to global stability

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Reference: *Clin Infect Dis 2007; 44:159-177 , Clin Infect Dis* 2013; 56:1445-1450, *Clin Infect Dis 2014; 59:S17-S75*

Problem statement

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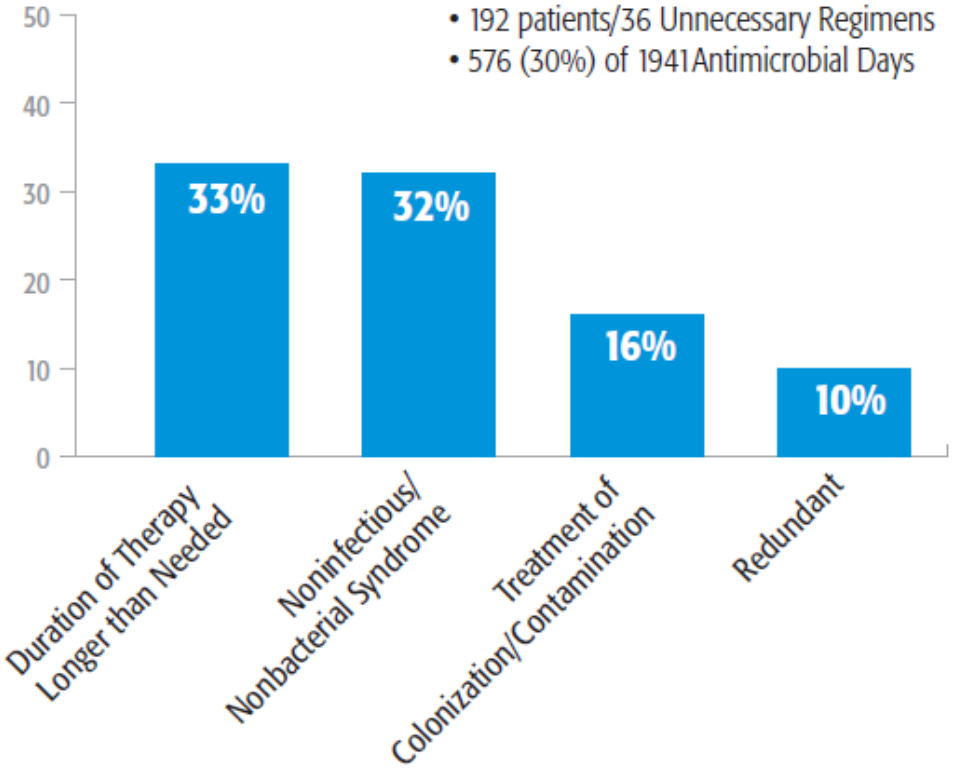
Antimicrobial Prescribing Facts: The 30% Rule

- ~ **30%** of all hospitalised inpatients at any given time receive antibiotics
- Over **30%** of antibiotics are prescribed inappropriately in the community
- Up to **30%** of all surgical prophylaxis is inappropriate
- ~ **30%** of hospital pharmacy costs are due to antimicrobial use
- **10-30%** of pharmacy costs can be saved by antimicrobial stewardship programs

[Hoffman et al., 2007; Wise et al., 1999; John et al., 1997]

Figure 2. "Unnecessary" Antimicrobial Therapy.



Adapted from Hecker MT. et al. Arch Intern Med. 2003;162:972-978.

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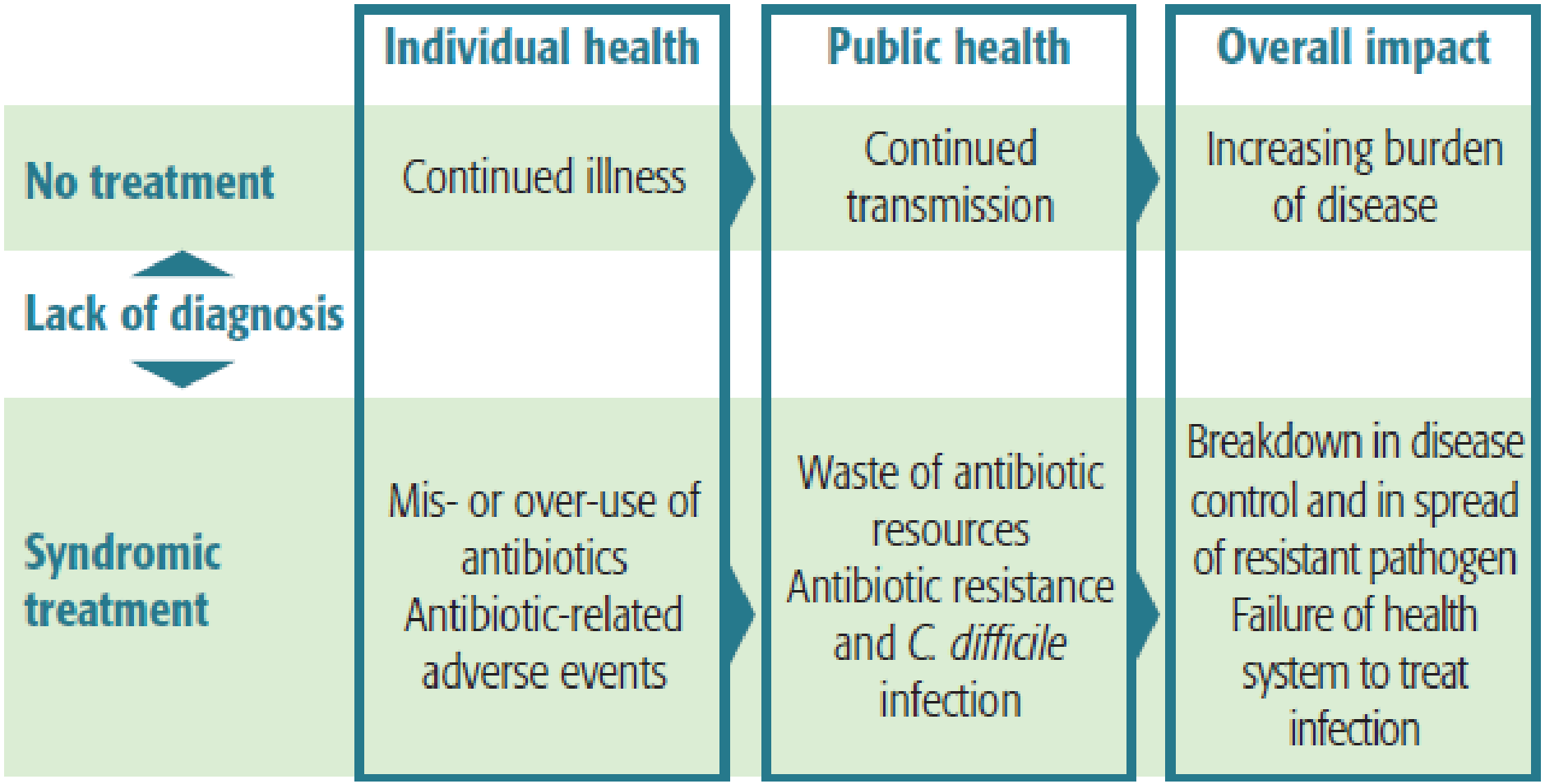
Figure 10. The high cost of poor diagnosis of infection.



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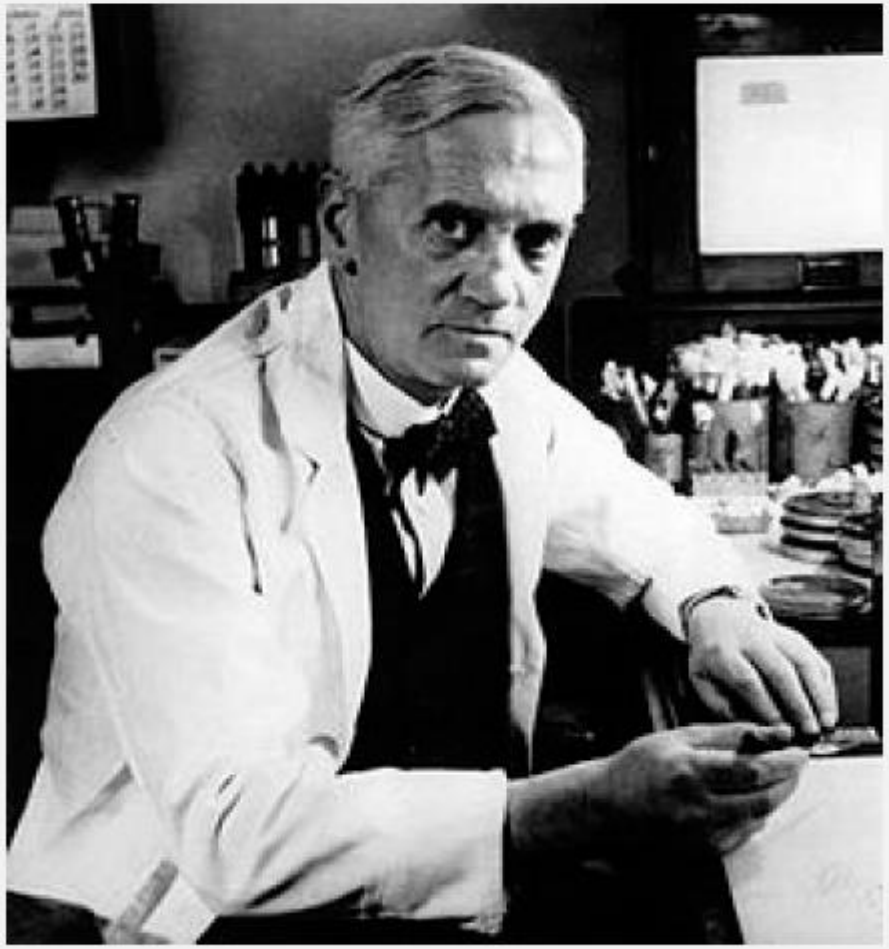
Birth of Antimicrobial Stewardship

"Microbes are educated to resist penicillin and a host of penicillin-fast organisms is bred out…

In such cases, the thoughtless person playing with penicillin is morally responsible for the death of the man who finally succumbs to infection with the penicillin-resistant organism. I hope this evil can be averted."



Fleming A. New York Times. 26 June 1945:21

Antibiotic Stewardship

"The right antibiotic
for the right patient,
at the right time,
with the right dose, and
the right route, causing
the least harm to
the patient and future patients"

www/cdc.gov/getsmart/healthcare/inpatient-stewardship

- Antimicrobial stewardship:
  is an **inter-professional effort**, across the continuum of care
  involves timely and optimal selection, dose and
- duration of an antimicrobial
  for the best clinical outcome for the treatment or
- prevention of infection
  with minimal toxicity to the patient
  and minimal impact on resistance and other
- ecological adverse events such as *C. difficile*"

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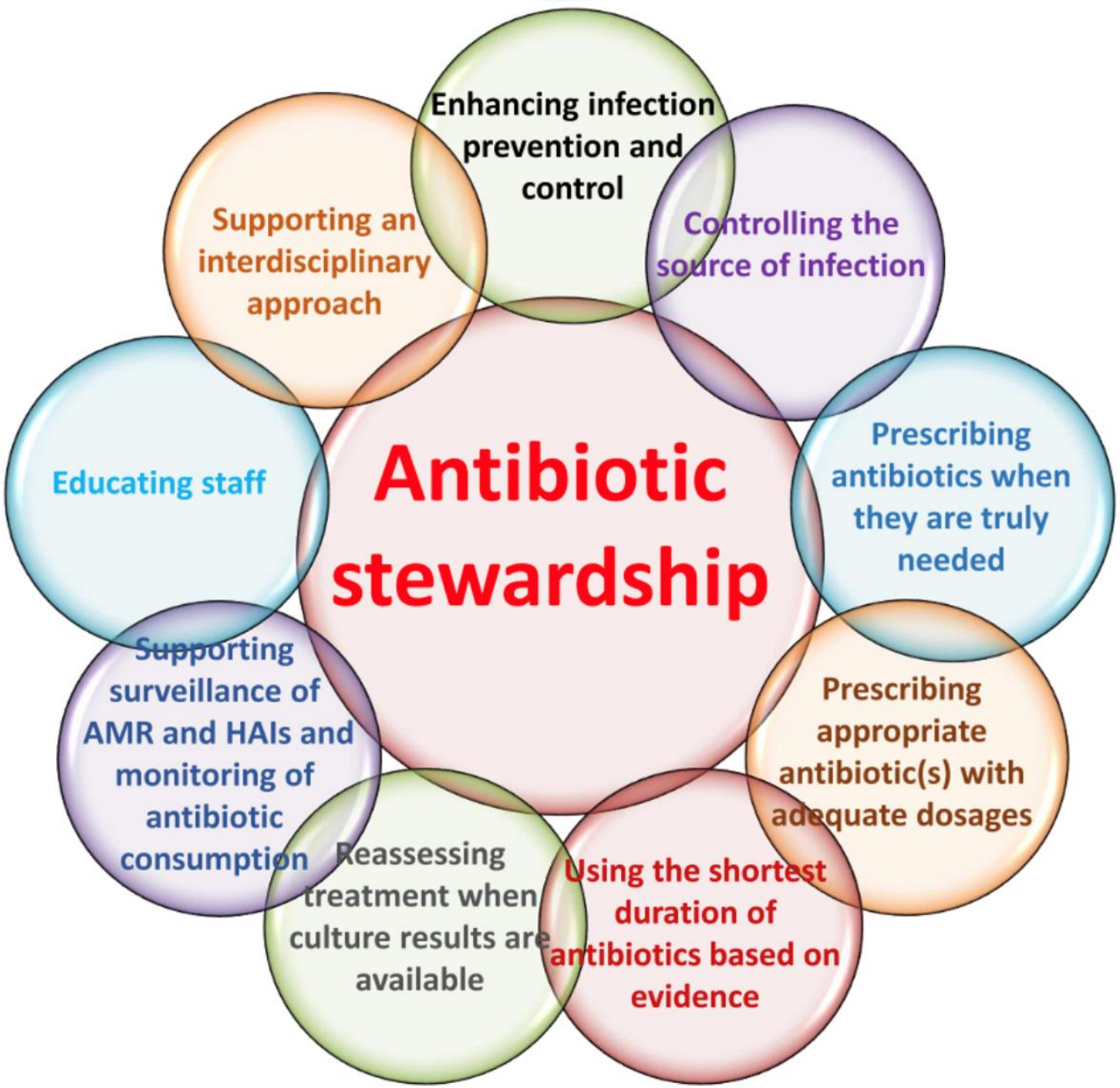

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Accurate and Rapid identification:
Diagnostic stewardship

The need for accurate antibiotics testing:

Each laboratory should develop, implement and regularly update SOPs that cover processing and storage, pathogen isolation, species identification and antimicrobial susceptibility testing (AST). AST should be conducted according to good laboratory practice including quality control at each stage and should meet an internationally-recognized performance and interpretive standard, such as those of the European Committee on Antimicrobial Susceptibility Testing (EUCAST)[10] and the Clinical and Laboratory Standards Institute (CLSI).[11]

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World Health Organization
Diagnostic stewardship is defined as: –

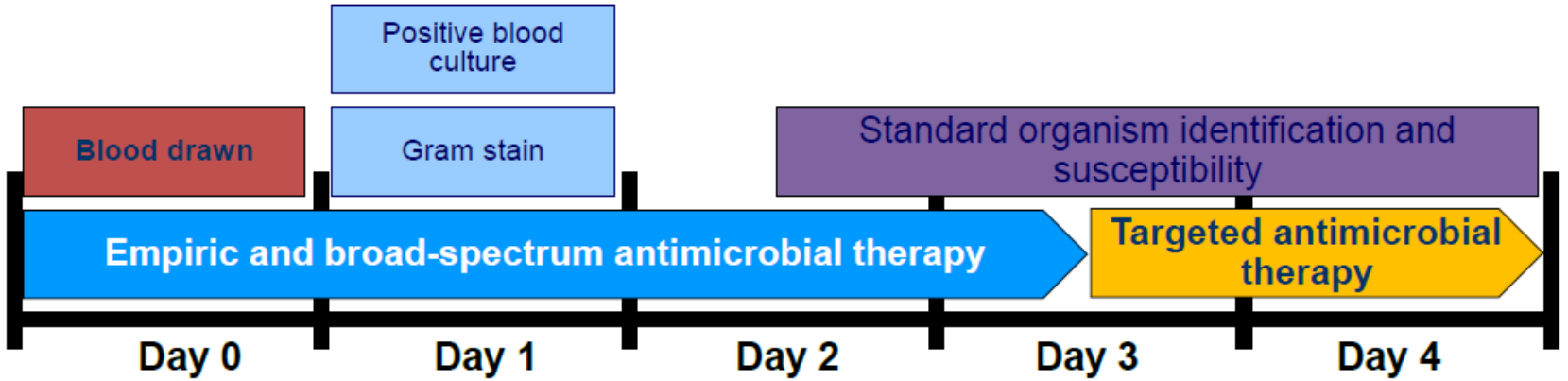
"coordinated guidance and interventions to improve appropriate use of microbiological diagnostics to guide therapeutic decisions. It should promote appropriate, timely diagnostic testing, including specimen collection, and pathogen identification and accurate, timely reporting of results to guide patient treatment ."

Microbiology laboratory today exceedingly pressured to perform
–exhaustive, expensive, clinically irrelevant [testing],"
–which, when misguided, "misleads physicians into erroneous diagnosis and inappropriate therapy"

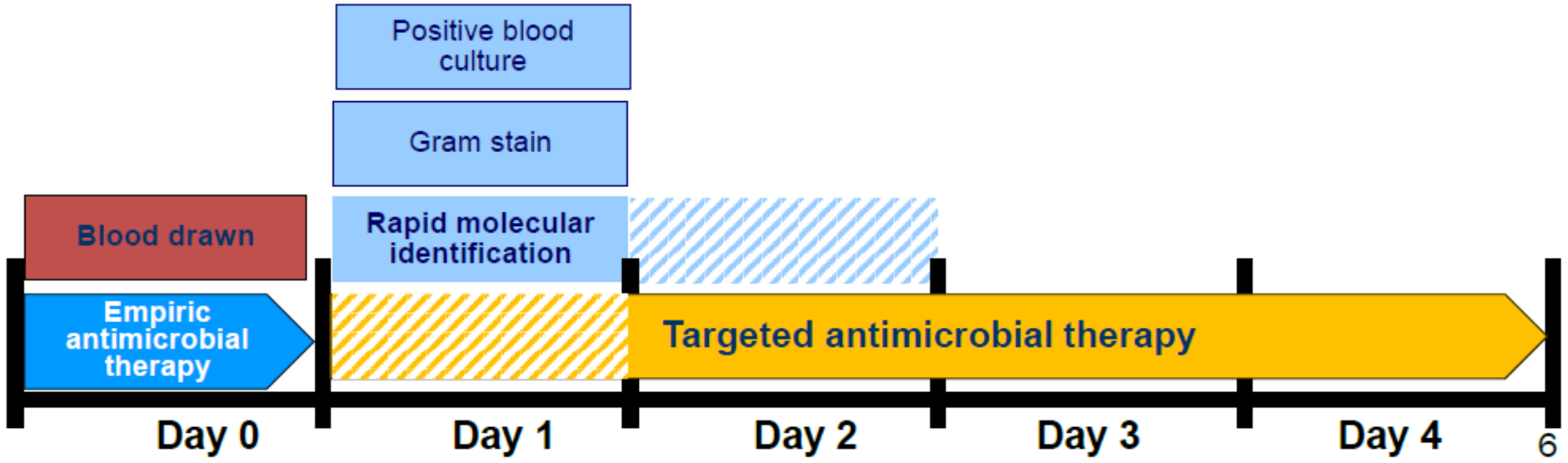
J ClinMicrobiol. 2017; 55:715–723.

Organism Identification and initiation of antimicrobial therapy

Traditional Identification & Testing Methods:



Rapid Molecular Identification Methods:



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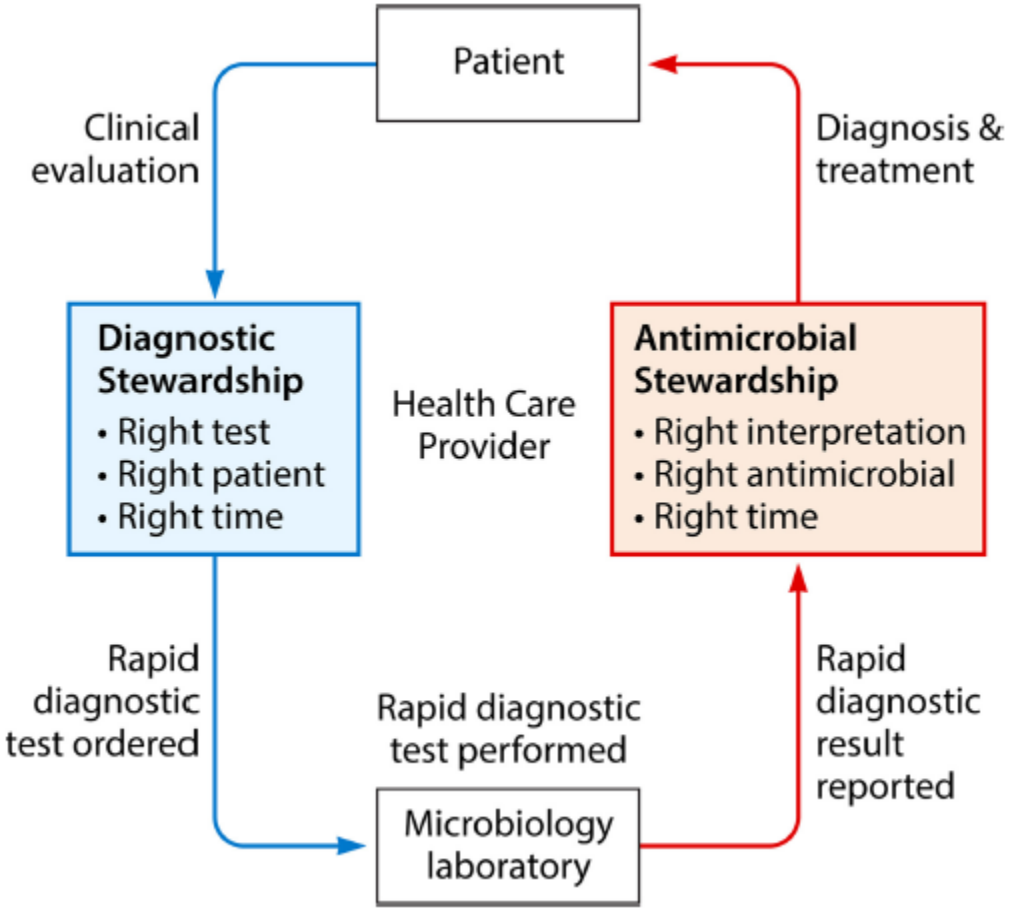
Kevin Messacar et al March 2017 Volume 55 Issue 3 Journal of Clinical Microbiology

Roles of diagnostic and antimicrobial stewardship in the implementation of rapid molecular infectious disease diagnostics in the clinical setting.



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Kevin Messacar et al March 2017 Volume 55 Issue 3 Journal of Clinical Microbiology

TABLE 1 Key diagnostic stewardship considerations for implementation of rapid infectious disease diagnostics

Goal	Key question	Key considerations and potential strategies
Right test	Is the test appropriate for the clinical setting?	Sensitivity and specificity Predictive values Testing volumes Diagnostic yield Laboratory feasibility Cost Clinical impact
Right patient	Will the clinical care of the patient be affected by the test result?	Laboratory test utilization committee Automatic laboratory reflex CPOE decision support Appropriate use criteria Indication selection Prior authorization Benchmarking Specimen rejection
Right time	Will the result be available in time to optimally affect care?	Time to specimen receipt Centralized vs point-of-care testing On-demand vs batched testing Specimen preparation time Run time Result reporting time

Table 7. The Golden Rules of Antimicrobial Prescribing "MINDME".

M	Microbiology guides therapy wherever possible
I	Indications should be evidence based
N	Narrowest spectrum required
D	Dosage appropriate to the site and type of infection
M	Minimise duration of therapy
E	Ensure monotherapy in most cases

Adapted from Antibiotic Expert Group. Therapeutic guidelines: antibiotic. Version 14. Melbourne: Therapeutic Guidelines Limited; 2010.

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Rapid Diagnostics: Current State

- Recent explosion of **FDA-approved rapid diagnostic testing (RDT)** methodologies for infectious diseases as the anti-infective pipeline remains stagnant

  - Major focus on **pathogens associated with increased morbidity, mortality, and excessive healthcare costs**
    Including influenza virus, methicillin-resistant *Staphylococcus aureus* **(MRSA)** vancomycin-resistant *Enterococcus* spp. **(VRE)**, *Clostridium difficile*, extended-spectrum β-lactamase **(ESBL)**- producing *Klebsiella* spp., ***Mycobacterium tuberculosis***, and ***Candida* spp.**

Polymerase Chain Reaction (PCR)-Based Testing

HOUSTON Methodist LEADING MEDICINE

Real-time PCR; Multiplex PCR

General method: Detection and amplification of a piece of target DNA using fluorescently labeled probes with primers

Overall Advantages	Overall Disadvantages
• Rapid results • Low detection limits • Specific organism detection or subtyping • Does not require growth on media • High throughput	• Susceptible to contamination • Require dedicated lab space for instruments • Sensitive to inhibitors present in many clinical specimens • Dependent on quality of nucleic axis primers and probes • Most require initiation from positive cultures/single colonies • Can not indicate viability of pathogen detected • Practical limitations can affect turnaround time

Afshari A, et al. Critical Care. 2012;16:222.
Bauer KA, et al. Clin Infect Dis. 2014;59 Suppl 3:S134-45.

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PCR-Based RDTs Available

HOUSTON Methodist LEADING MEDICINE

	Organisms/ Antibiotic Resistance Targets	Detection Time, hours	Manufacturer	Test Commercial Name
Real-time PCR	MRSA	2	Roche Diagnostics	Light Cycler MRSA
	C. difficile	2	BD GeneOhm	BD GeneOhm Cdiff Assay
		3	Gen-Probe Prodesse	ProGastro Cd Assay
Multiplex PCR	MSSA, MRSA, CoNS	2	BD GeneOhm	BD GeneOhm Staph SR
		1	Cepheid	Xpert MRSA/SA BC
	MSSA, MRSA	1	Cepheid	Xpert MRSA/SA SSTI
	S. aureus, Staphylococcus epidermidis, Streptococcus spp. *E. faecalis, E. faecium, Micrococcus* spp, *Listeria* spp, mecA, vanA, vanB	2.5	Nanosphere	Verigene: BC-GP
	C. difficile	0.5	Cepheid	Xpert C. difficile
		0.75	Cepheid	Xpert C. difficile/Epi
	E. coli, K. pneumoniae, K. oxytoca, P. aeruginosa, Serratia marcescens, Acinetobacter spp, *Proteus* spp, *Citrobacter* spp, *Enterobacter* spp, KPC, NDM, CTX-M, VIM, IMP, and OXA genes	<2	Nanosphere	Verigene: Gram-negative blood culture
	Multiple bacterial, fungal, viral pathogens, and mecA, vanA/B, carbapenem resistance	1	BioFire Diagnostics	FilmArray System & panels

Blood Culture Systems

bioMerieux BacT/Alert

BD BACTEC

Thermo VersaTREK

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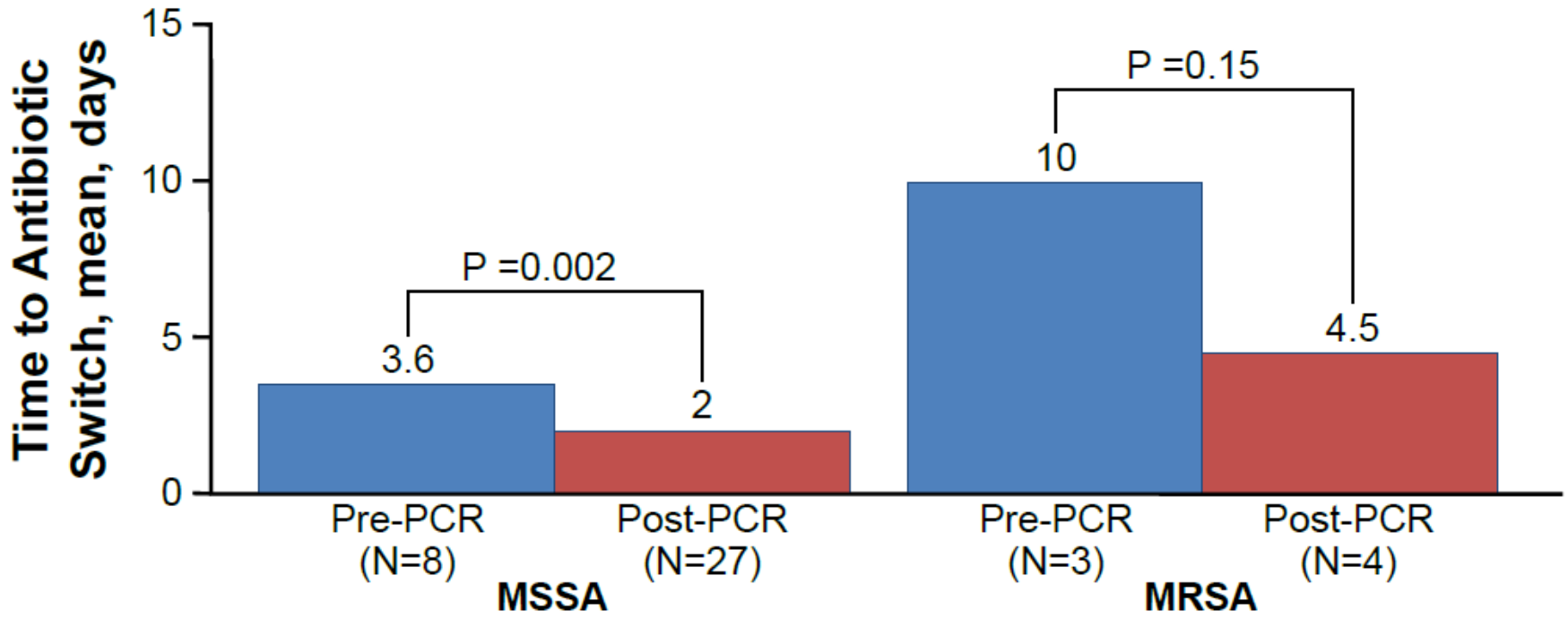
PCR-Based Testing – Clinical Outcomes

HOUSTON Methodist LEADING MEDICINE

Rapid *S. aureus* Identification and Targeted Antimicrobial Therapy in Patients with *S. aureus* Bacteremia

Single-center, Non-equivalent, Comparative Study [2008, 2009]



PCR results were communicated to an ID pharmacist who recommended effective, targeted antimicrobial therapy and an ID consult

Bauer KA, et al. Clin Infect Dis. 2010;51(9):1074-1080.

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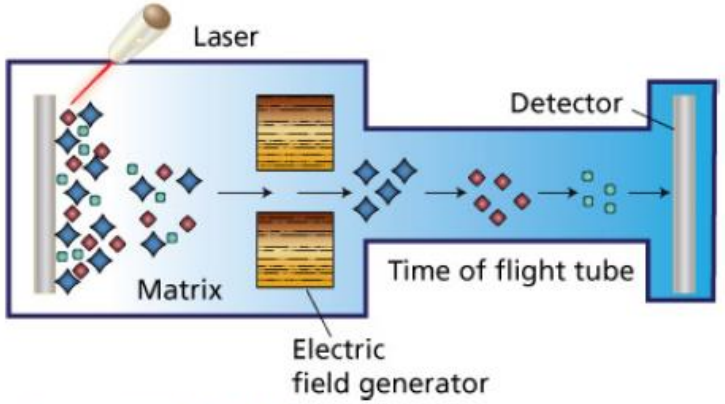
Matrix-Assisted Laser Desorption/Ionization Time of Flight (MALDI-TOF) Mass Spectrometry

- ID of bacteria, yeast and fungi
- Identification by *signature* high abundance proteins (primarily **ribosomal proteins**)
- Signature protein patterns are compared / matched to an extensive open database

- Rapid identification of bacteria, yeast and fungi
  - Minimum of 24-96 h reduced turnaround time compared to conventional culture-based methods.
  - Even more dramatic results for certain organisms (i.e. *Legionella*).
- Test directly from positive blood cultures
  - Bypasses time-consuming subculture steps



Wimmer JL, J Clin Microbiol. 2012;50:2452-4.
Seng P. Clin Infect Dis. 2009;49:543-51.

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Azole Resistance in *Aspergillus fumigatus*

- Of particular concern are resistant *A. fumigatus* isolates carrying either TR34/L98H or TR46/Y121F/T289A genetic resistance markers, which have been associated with environmental triazole fungicide use rather than previous patient exposure to antifungals
- More resistance cases in Europe
- Most labs don't perform mold susceptibility testing
- TAT can be reduced by MALDI-TOF prcotocols

https://www.cdc.gov/fungal/aspergillus-resistance.html

Rapid Diagnostic Tests & Antibiotic Stewardship

- Rapid Diagnostic Tests are **“game changing” for patient care** moving forward Advances in RDTs provide new opportunities for stewardship programs
- Enhance function of clinical microbiology laboratories
- Yields **significant improvement to patient care**, increases the effectiveness of ASPs and infection control efforts
- Represents one of the few bright spots in the changing world of escalating antimicrobial resistance and stewardship

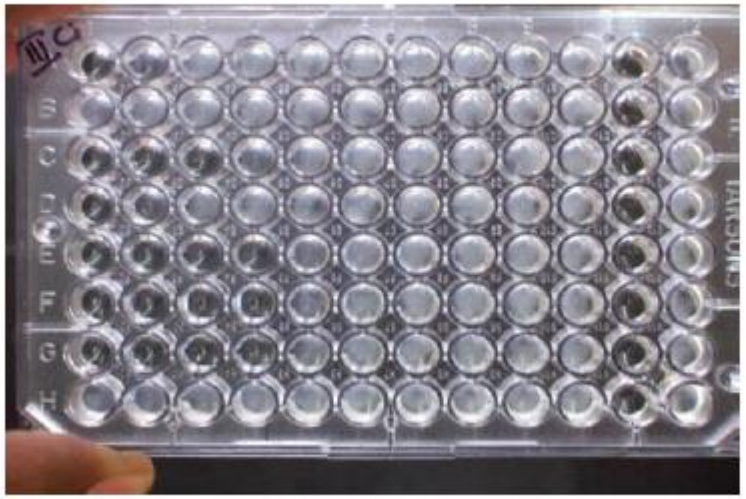
Ideal Molecular Test

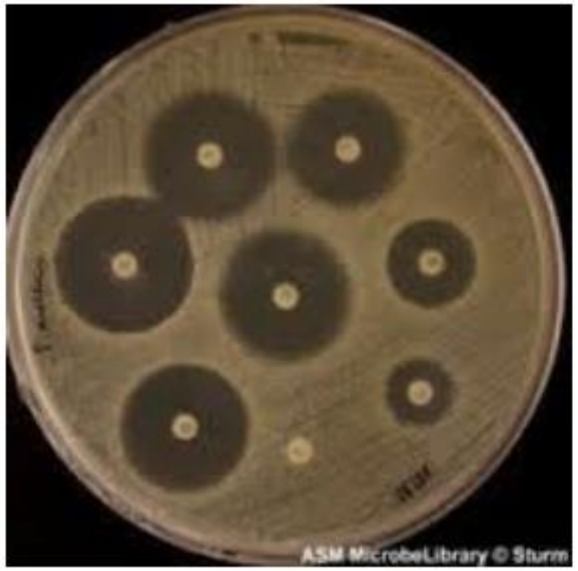
Key questions to ask about any new molecular test:

- Does it provide a clinically useful result?
- Does it cover your routine organisms of interest?
- Does it provide useful, reproducible results?
- Is it cost effective?
- Does it provide good or improved turn-around-time?

Manual AST

- Three Common Methods
  - Broth Microdilution (BMD)
  - Disc Diffusion (DD)
  - Etest





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Manual AST – disc diffusion Workflow

1

Select isolated bacterial colonies

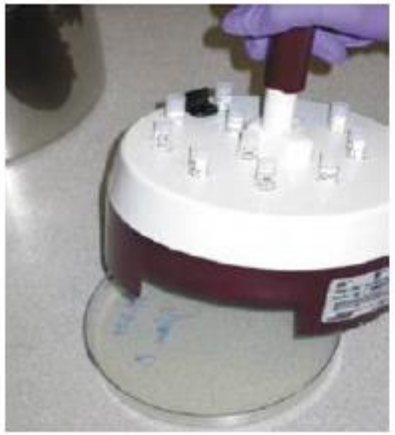
2

Make a 0.50 McFarland (Bacterial suspension)

3

Inoculate the agar plate

4

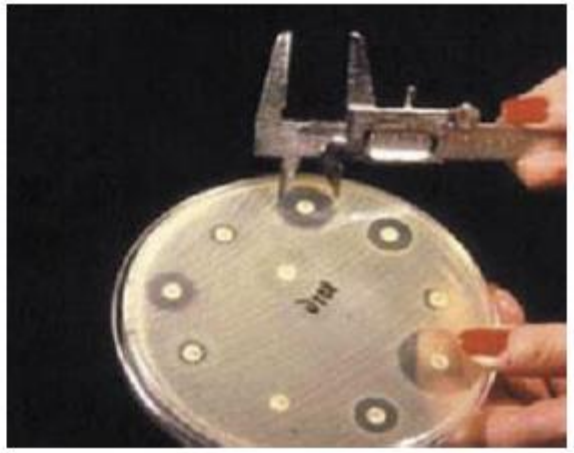


Place discs containing antibiotic on plate

5

Incubate the agar plate overnight

6



Measure the zone of inhibition

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Automated AST– VITEK workflow

Select isolated bacterial colonies

Make a 0.50 McFarland (Bacterial suspension)

Add some of the McFarland solution to another vial

Filled with special saline

VITEK AST Card

VITEK ID Card

VITEK Instrument

Hands-on Time: 10-20 minutes
Time to Results: 12-18 hours

Automated AST – BD Phoenix Workflow

Select isolated bacterial colonies

Make a 0.50 McFarland (Bacterial suspension) In BD Phoenix ID Broth

Add some of the prepared ID Broth solution to AST Broth

Add drop of indicator fluid to BD Phoenix AST Broth before adding ID Broth solution

Inoculate panel with ID Broth

Inoculate panel with AST Broth

BD Phoenix Panel

BD Phoenix Instrument

Hands-on Time: 10-20 minutes
Time to Results: 12-18 hours

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RT-PCR, Multiplex, Microarray: Sample-to-answer technology

- MTB + rifampin R
- *C. difficile* toxin genes
- Bacterial resistance genes
  - mecA, vanA, carbapenemases
- Microarray/multiplex panels
  - Respiratory
  - CNS infection
  - Gastroenteritis
  - Bloodstream infection (**+ culture**)

The impact of rapid molecular diagnostic testing for respiratory viruses on outcomes for emergency department patients

Nasir Wabe, Ling Li, Robert Lindeman, Ruth Yimsung, Maria R Dahm, Kate Clezy, Susan McLennan, Johanna Westbrook and Andrew Georgiou

Med J Aust 2019; 210 (7): 316-320. || doi: 10.5694/mja2.50049

Published online: 8 April 2019

Question : whether Rapid polymerase chain reaction (PCR) testing for influenza and respiratory syncytial viruses (RSV) in emergency departments (EDs) is associated with better patient and laboratory outcomes than standar multiplex PCR testing ?

1491 consecutive patients tested by standard multiplex PCR during July–December 2016, and 2250 tested by rapid PCR during July–December 2017.

Main outcome measures: Hospital admissions; ED length of stay (LOS); test turnaround time; patient receiving test result before leaving the ED; ordering of other laboratory tests.

Results: Compared with those tested by standard PCR, fewer patients tested by rapid PCR were admitted to hospital (73.3% v 77.7%; P < 0.001) and more received their test results before leaving the ED (67.4% v 1.3%; P < 0.001); the median test turnaround time was also shorter (2.4 h [IQR, 1.6–3.9 h] v 26.7 h [IQR, 21.2–37.8 h]). The proportion of patients admitted to hospital was also lower in the rapid PCR group for both children under 18 (50.6% v 66.6%; P < 0.001) and patients over 60 years of age (84.3% v 91.8%; P < 0.001). Significantly fewer blood culture, blood gas, sputum culture, and respiratory bacterial and viral serology tests were ordered for patients tested by rapid PCR. ED LOS was similar for the rapid (7.4 h; IQR, 5.0–12.9 h) and standard PCR groups (6.5 h; IQR, 4.2–11.9 h; P = 0.27). Conclusion: Rapid PCR testing of ED patients for influenza virus and RSV was associated with better outcomes on a range of indicators, suggesting benefits for patients and the health care system. A formal cost–benefit analysis should be und

The BioFire® FilmArray® Torch

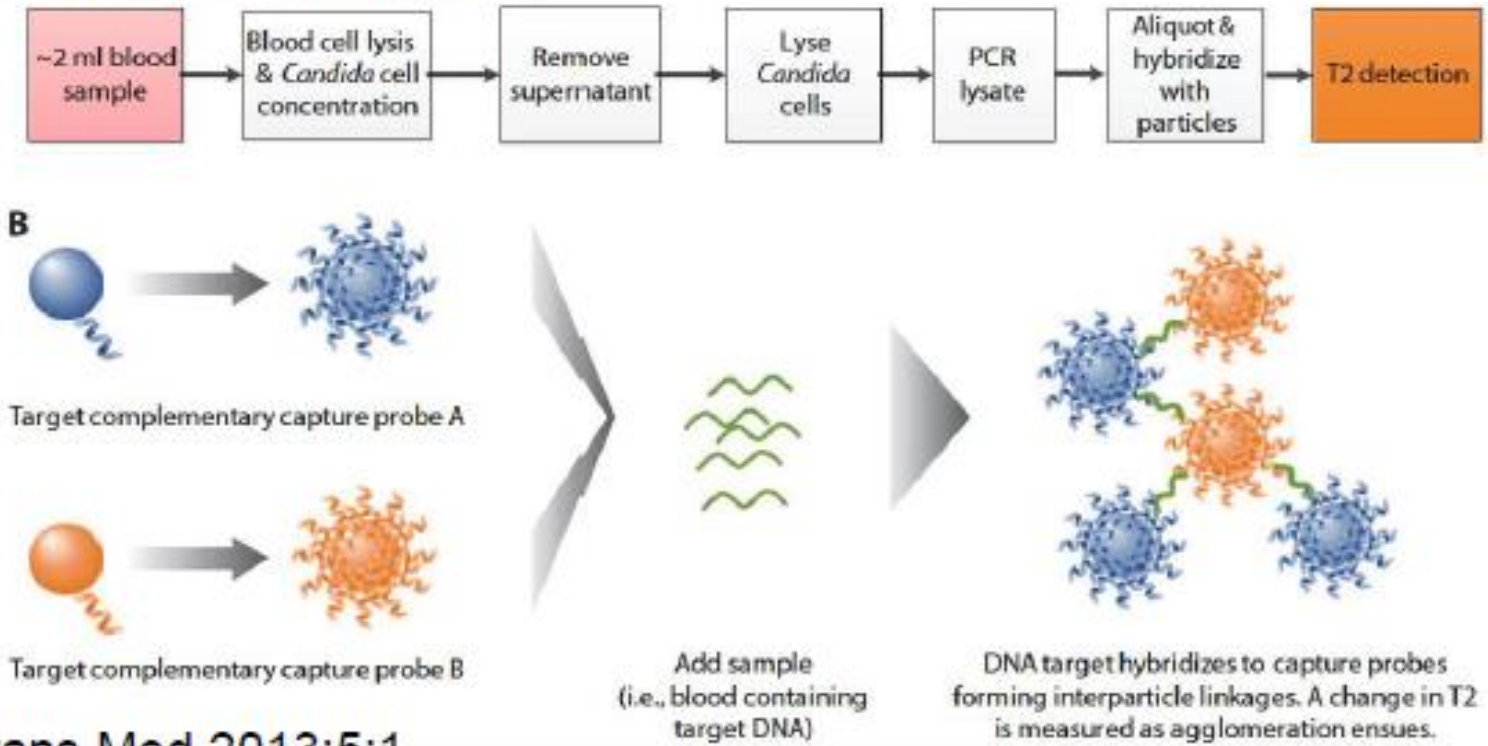
- latest advancement in molecular infectious disease diagnostics.
- High throughput, fully integrated, random access system
- sample preparation, reverse transcription-PCR, PCR, and detection in a freeze-dried format in a pouch
- The Film Array platform is a closed diagnostic system allowing high-order multiplex PCR analysis with automated readout of results directly from positive blood cultures in 1 h.
- Film Array blood culture identification (BCID) panel, includes 19 bacteria, five yeasts, and three antibiotic resistance genes.

- The Film Array could identify microorganisms in 153/167 (91.6%) samples with monomicrobial growth.
- Thirteen of the 167 (7.8%) microorganisms were not covered by the Film Array BCID panel.
- In 6/167 (3.6%) samples, the Film Array detected an additional microorganism compared to blood culture.
- When polymicrobial growth was analyzed, the Film Array could detect all target microorganisms in 17/24 (71%) samples.
- Twelve blood culture bottles that yielded a positive signal but showed no growth were also negative by Film Array.

Ashok Rattan., et al. “Laboratory Investigations for Neonatal Sepsis". Acta Scientific Microbiology Special Issue 1 (2019): 42-46.

Magnetic resonance diagnostics

- Magnetic particles coated with capture probes
- Amplified product in sample causes agglomeration
- Detected with MR signal



Neely, et al. Sci Trans Med 2013;5:1.

T2MR diagnostics

- T2 Magnetic Resonance (T2MR) is a miniaturized, **magnetic resonance based diagnostic approach** that measures how water molecules react in the presence of magnetic fields.

- Advantage: Applied directly to blood sample
- –Time to detection and species ID ~4 hours
- Bacterial panel FDA approved

Mylonakis E, et al. Clin Infect Dis 2015;60:892.

Why T2?

"This technology has shown it can expedite the detection of candidemia. As a result, patients receive more prompt and appropriate antifungal therapy." – Rachel Kenney, PharmD, Henry Ford study co-author

Faster Targeted Therapy: The median time to appropriate antifungal therapy was reduced from 39 hours to 22 hours (P=0.003)

Patients tested with the T2Candida were appropriately treated in a median of 5 hours, compared to 44 hours with blood culture[1]

Reduced Length of Stay: Henry Ford reported a reduced median ICU length of stay by 7 days and an overall 4 day reduction[1]

Improved Patient Outcomes: Reduction in occurrence of ocular candidiasis cases were (P<0.28)

1. Wilson N, et al. **Journal of Antimicrobial Stewardship**, 2017

Invasive Candidiasis

- Antifungal resistance is a particular problem with *Candida* infections.
- Some types of *Candida* are increasingly resistant to the first-line and second-line antifungal medications, such as fluconazole and the echinocandins (anidulafungin, caspofungin, and micafungin).
- Increase resistance *Candida glabrata* and *Candida krusei*
- Multidrug-resistant *Candida* infections (those that are resistant to both fluconazole and an echinocandin)
- Emerging antifungal resistance has been identified in species like *Candida auris*.

Future Medicine

JOURNALS BOOKS ABOUT US CONTACT US

FUTURE MICROBIOLOGY, VOL. 13, NO. 10 | REVIEW

Use of T2MR in invasive candidiasis with and without candidemia

Ioannis M Zacharioudakis, Fainareti N Zervou & Eleftherios Mylonakis

Published Online: 24 May 2018 | https://doi.org/10.2217/fmb-2018-0079

The Molecular Lab – Labs of the Future









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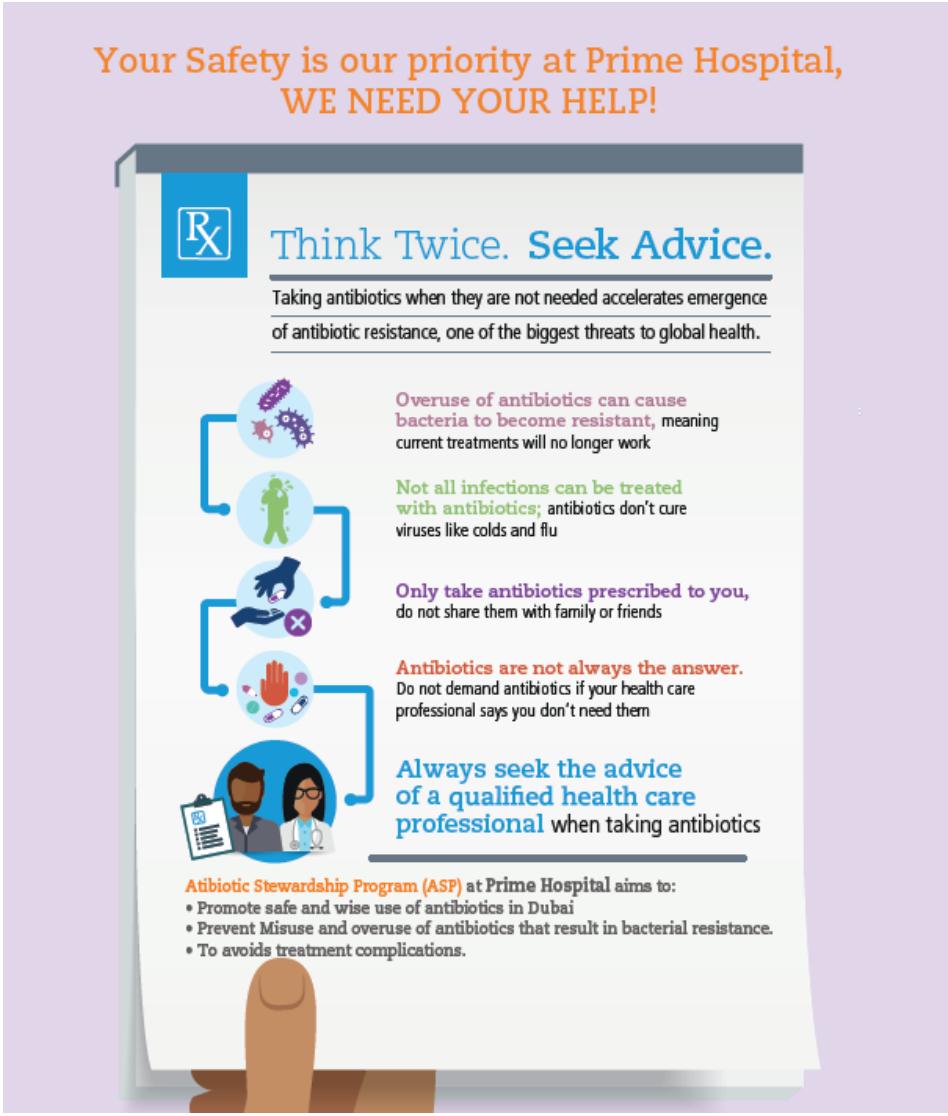
Newer technologies

- Current technologies are excellent at affirmatively answering the questions of whether the patient is infected and, if so, with what, by identifying organisms
- Directly from specimens (Biofire, T2MR)
- Using syndromic panels
- MALDI-TOF MS, and next-generation sequencing
- However, technology currently cannot, and perhaps **will never be able to, provide a definitive negative result to rule out infection.**
- The ability of rapid diagnostic technologies is to answer the question of what will treat the infection ?

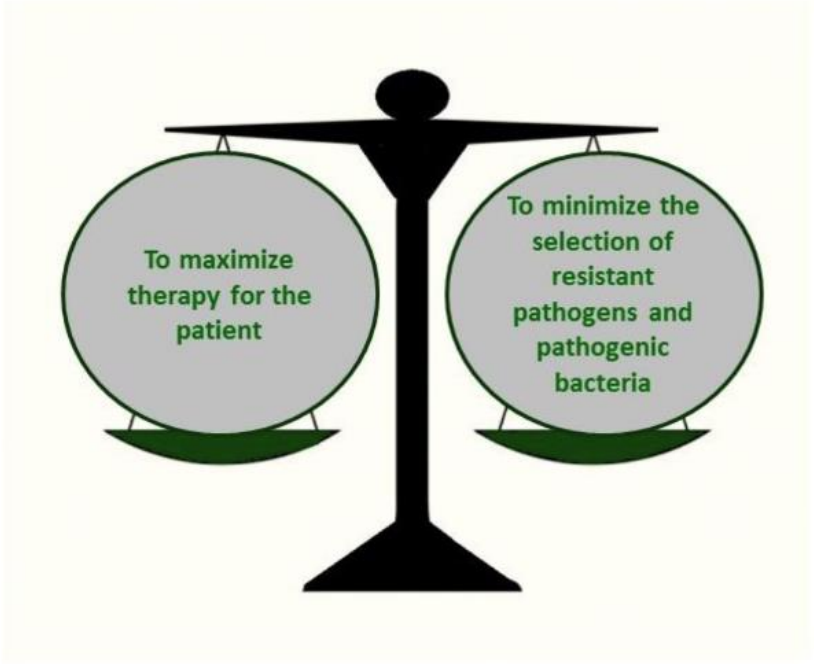
Education and awareness



- Awareness among all health care workers by conferences and workshops
- Parents
- school children
- colleges
- TV shows
- Facebook
- Radio talks

Antibiotics use: finding the right balance

Global Alliance for Infections in Surgery



On one hand clinicians should offer optimal therapy for the individual patient under their care; on the other hand they should limit the impact of the antibiotic in order to prevent the selection of resistant pathogens and pathogenic bacteria such as *C. difficile*.

ACT NOW

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Battle Against Antimicrobial Resistance Requires Teamwork

- Requires team work - more stakeholders involved in ASP:
  - Infection Prevention
  - Infectious Disease
  - Pharmacy
  - Physicians – ICU, ED especially
  - Nurses – ICU
  - Board of Directors - educated
  - Diagnostic Laboratory Directors
  - C Suite: CEO, CNO, CMO, CFO – for financial resources and support
  - Microbiology Lab

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UAE Antibiotic Stewardship Program & AMR team

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