

Rapid Diagnostic Tests and Early Reporting

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Bloodstream infections: opportunities for outcome improvement

Disclosures

- Lectures for
 - Abbott Diagnostics (Sweden)
- Research projects in collaboration with
 - BD/Kiestra
 - Q-linea
 - Luminex

The patient with sepsis

- Comes to the ER 24/7/365
- Is severely ill!
- Will benefit from rapid institution of antibiotics

But!

- Increasing antibiotic resistance puts emphasis on tailored therapy
- The patient does not always arrive at the "correct" hospital
- The personnel do not always have the incentives to handle the samples correctly
- The laboratory is not always open 24/7/365

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WHAT MAKES A RAPID TEST RAPID?

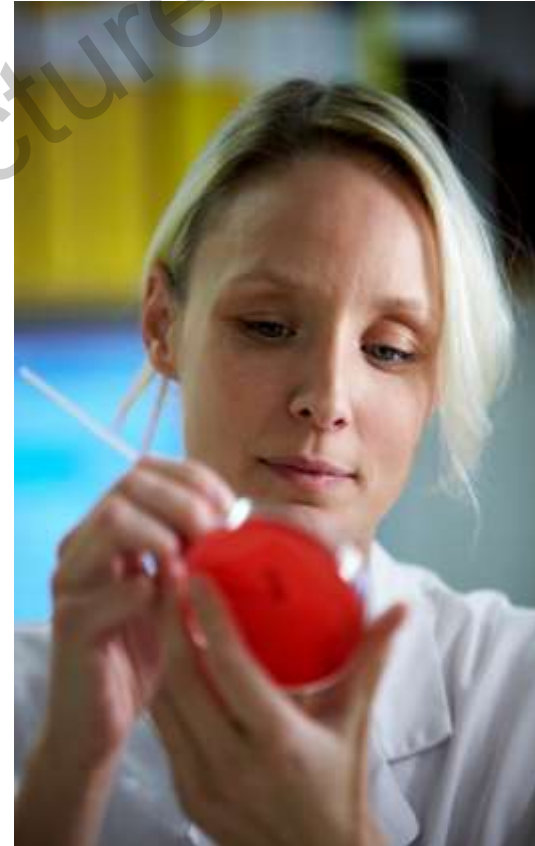
Pre analysis

- Transports
- Knowledge
- Opening hours
- External Blood-culture cabinets
- Decentralized ER laboratories
- Electronic referrals



In the lab

- Opening hours
- Dedicated staff
- Knowledge
- Prioritize the diagnosis of sepsis
- Reports
 - Electronic
 - Transparent
 - Preliminary
 - Telephone



In the ward

- Electronic chart systems
- Knowledge
- Consultants available for instant discussion regarding antibiotic therapy



What you can do on monday

- Make your automated blood culture system available for the introduction of vials 24/7/365
(van der Welden 2010, Schneiderhan 2013)
- "Active culturing"



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WHAT IS AVAILABLE IN SEPSIS DIAGNOSTICS?

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Tests in the ER

- History of the patient!
- Clinical Investigation

- CRP
- PCT
- WBC, neutrophils
- Lactate
- Etc...

Dynamic tests – To be followed over time!

BLOOD CULTURE

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Automated Blood-culture systems

- Aerobic and anaerobic culture
 - Pediatric, Mycosis?
- Several systems
 - BacTAlert (Biomérieux)
 - BacTec (BD)
 - VersaTrek (ThermoFischer)



Rapid ID from positive BC vials (1)

- PCR for a range of pathogens
 - The most common pathogens
 - Some resistance genes
 - 2-6h to ID
- Broad range PCR
 - At least 2-6 hours to ID
 - Needs sequencing
- Quick-FISH (PNA-FISH)
 - Gram stain 10 min
 - Quick-FISH 20 min
 - The most common pathogens in smaller panels (Calderaro CMI 2013)



Rapid ID from BC vials (2)

- MALDI-TOF (direct method)
 - 10-30 min
- “Active culturing” and MALDI-TOF
 - Rapid incubation on nutritious agar in different environments
 - 3 hours at its best to species ID

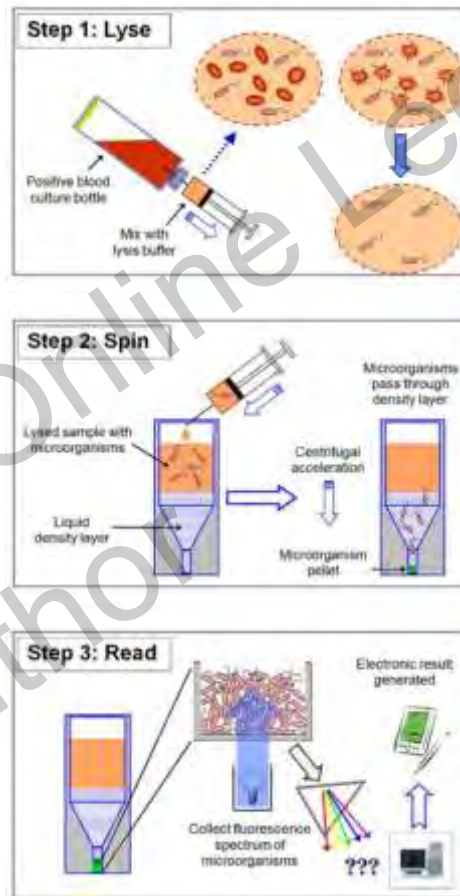


MALDI-TOF and Sepsis

- Positive Blood culture
- Centrifugation and extraction steps (10-30 min)
- Lower limit for genus/species ID has been suggested.
- ID obtained in 66 - 99% (Martiny CMI 2012, Lagacé-Wiens 2012, Klein 2012, Machen PLoS One 2014, Schieffer JAM 2014)
- Impact of Rapid ID
 - Time to species ID 84 → 56h
 - time to effective tx 30 → 20h
 - time to optimal tx 90 → 47h (Huang CID 2013)
 - Change of treatment in 13% of adults.
 - 37% helpful in peadiatrics as the result showed contamination. (Martiny CMI 2013)
 - 58h earlier report if 24/7 introduction of BC vials and MALDI 24/7. (Schneiderhan Clin Chem 2013)

In pipeline

- Intrinsic fluorescens



But don't forget!

- Gram stain
- Agglutination tests
- Rapid coagulase test
- Other culture results!
- And microbiologistics!!



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SPECIES ID DIRECTLY FROM BLOOD

ID before enrichment

Antigentests

- Pneumococci (urine, CSF)
- Legionella (urine)
- Meningococci (CSF)

- Limited sensitivity
- Acceptable specificity
- Predictive value?

Nucleic acid detection

- SeptiFast, Roche
- Sepsitest, Molzym
- Magicplex, Seegene
- Vyoo, Bionity
- Inhouse methods

- Limited sensitivity
- Acceptable specificity
- Predictive value?

NAT directly from blood

Benefit

- Could decrease time to Species ID
- Less problem with insignificant pathogens
- Limited Species-panels
- Can detect bacteria after ab-tx is initiated
- Could be of value for "Rule-In" diagnostics

Draw back

- Low Sensitivity!
 - 1 – 1.5 mL whole blood
- Will be batched → reduce the speed.
- Needs trained personell.
- Sometimes a second sequencing step is needed.

In pipeline

PlexID (Abbott)

- Lysis and Extraction 2h
- PCR 2h
- ESI-MS and interpretation
70 min
- 6-8h

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SUSCEPTIBILITY TESTING IN SEPSIS

General aspects of AST

- Resistance detection \neq Susceptibility testing
- So far no techniques with high sensitivity for detection of resistance mechanisms directly from blood sample
- All commercial and/or molecular systems are less dynamic to local epidemiology

Molecular techniques

- Detects well known resistance mechanisms
- Well validated for MRSA
- Some panels for Carbapenemases and ESBLs
- Some specific assays for VRE

Phenotypic AST

- Susceptibility testing
- DD or Gradient strip
 - 6-8h
(Jonasson et al Poster P0333 and PO334 ECCMID 2014, Sundqvist et al, manuscript, Eurostar project)
- Automated
 - 16h (5-16h)
(Wimmer JCM 2012, Machen PLoS One 2014)



Disk Diffusion

- Directly from BC-vials (Jonasson et al P0334)
 - Non-standardised inoculum
 - Over night incubation or short as below
- Eurostar Rapid Disk
 - EUCAST methodology (www.eucast.org)
 - BUT! Short incubation time.
 - Enterobacteriaceae (6h)
 - Pseudomonas, Enterococci, *S.aureus* (8h)
 - *Haemophilus influenzae* and *Pneumococci* (8h)

In pipeline

- MALDI-TOF (Jung et al EJCMI 2013)
- Microcalorimetry (Braissant et al JCM 2014)
- Combined Broth Dilution and PCR → AST

Future perspectives

- Molecular based detection from whole blood
 - Black box
 - Random access
 - Affordable
 - If accomplished: preliminary report in 6-8h?
- Culture (usually positive within 20h)
 - MALDI-TOF + rapid DD → definitive report in <30h?

Future perspectives

- Microbiology lab
 - Open 24/7/365
 - Skilled staff 24/7/365
 - Don't forget other samples from the critically ill patient
 - Take control of the transport organization and the referrals!

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