Rapid Diagnostics to support Antimicrobial Stewardship

2015
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Disclosures

- Consulting or lecture honoraria from Bayer, Wyeth, Janssen-Cilag, Pfizer, Astra Zeneca, Cubist, Merk
- Investigator on antibiotic trials for Bayer, Pfizer, Basilea, Wyeth, Astra Zeneca
- Investigator of Surgihoney Reactive Oxygen, and Momentum Cognitor minus.
- Recent General Secretary of British Society of Antimicrobial Chemotherapy

Is rhetoric enough?

Bad Bugs Need Drugs

10x'20

Ten new ANTIBIOTICS by 2020

ANTIBIOTIC ACTION

THE ARMS RACE

"Antibiotic resistance - one of the three greatest threats to human health."

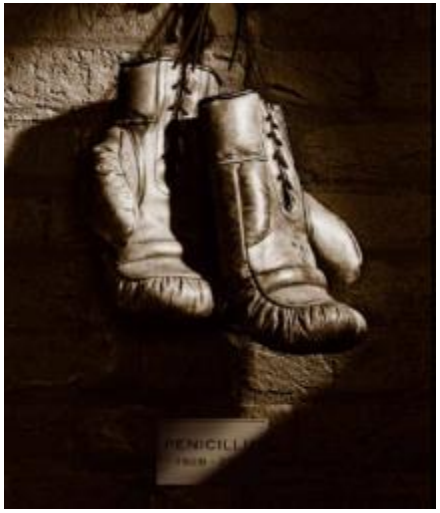
World Health Organisation, 2009

PENGUIN SPECIALS

THE DRUGS DON'T WORK

A GLOBAL THREAT

PROFESSOR DAME SALLY C. DAVIES



Review on Antimicrobial Resistance

Antimicrobial Resistance: Tackling a crisis for the health and wealth of nations

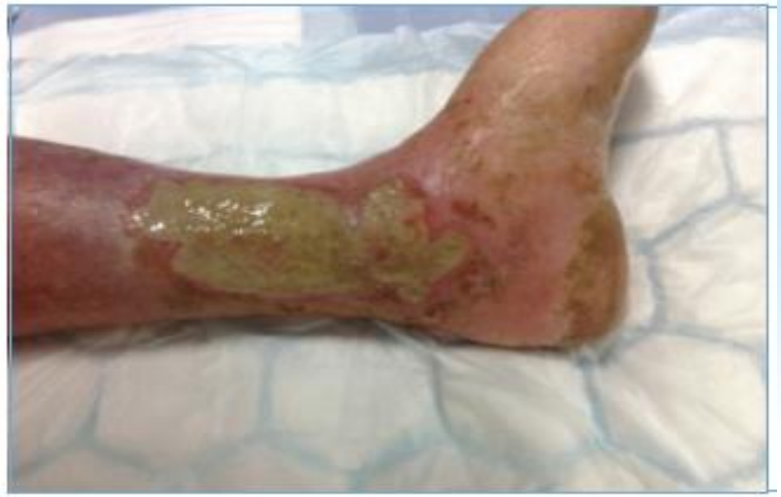
The Review on Antimicrobial Resistance
Chaired by Jim O'Neill
December 2014

Lateral thinking required

- **Rapid diagnostics**
  - **To ensure antibiotics given when bacterial infection present**
  - **To stop when infection not present**
  - **To rationalise / deescalate**
- **Infection prevention**
  - **To reduce need for antibiotics**

Infection prevention

- Standard IPC
- Vaccination
  - Pfizer staphylococcal vaccine projects
- Non-antibiotic prevention therapies
  - Surgihoney / Reactive oxygen species
  - Reduces bacterial bioburden and biofilm in wounds, ulcers, ?resp tract, uroepithelium
  - Surgical prophylaxis

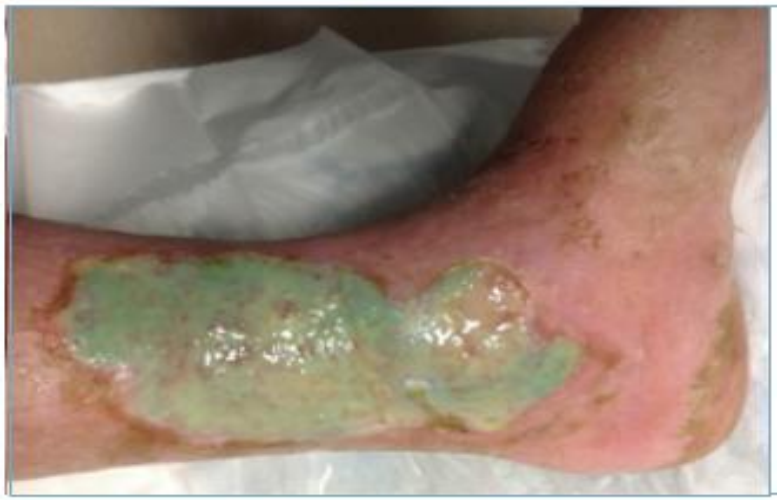


Day 1

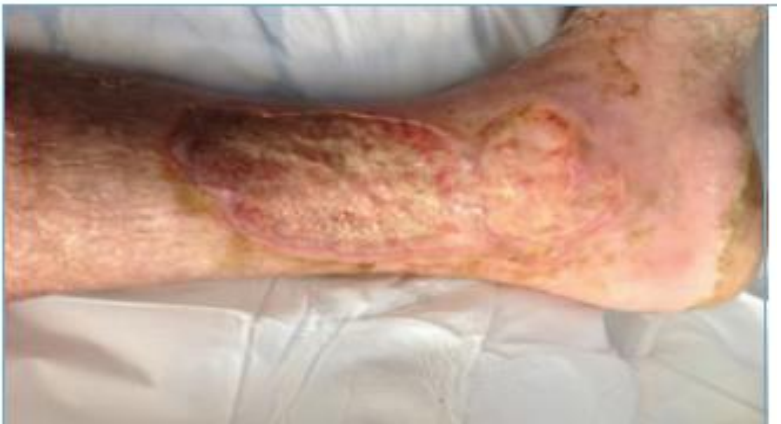
Clearance of MDR colonisers

Reduction in SSI

Huge potential in mucosal biofilm and colonisation.



Day 4



Day 10

Reactive Oxygen™

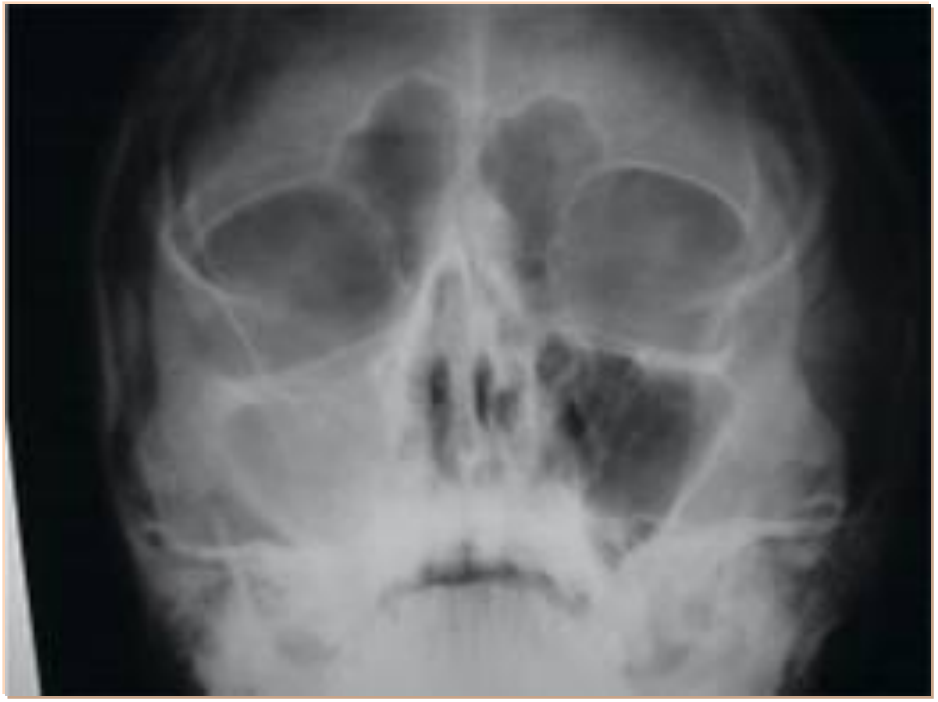
Surgihoney™

The World's Most Powerful, Non Toxic, Topical Antimicrobial

Future developments

Nebulised RO in COPD/bronchiectasis

Chronic sinusitis

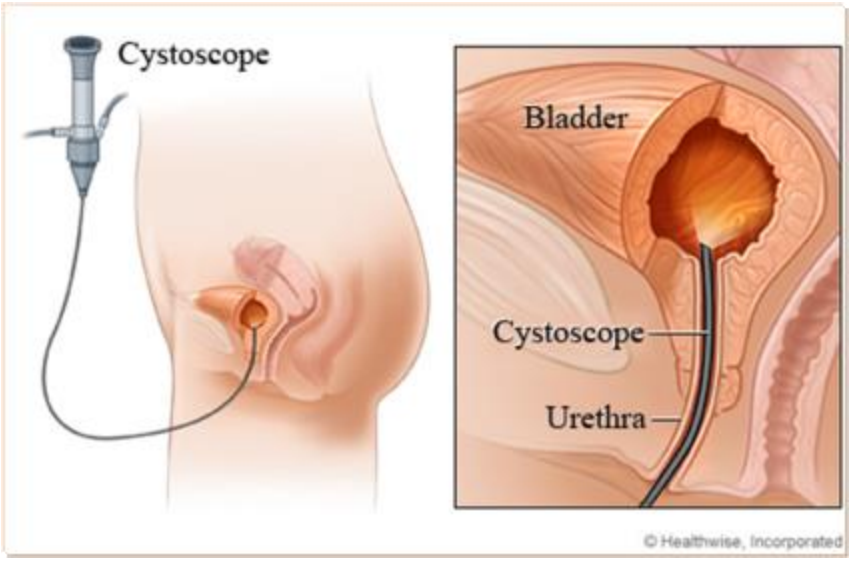


Reactive Oxygen™

Reactive Oxygen™

Complex joint replacement surgery

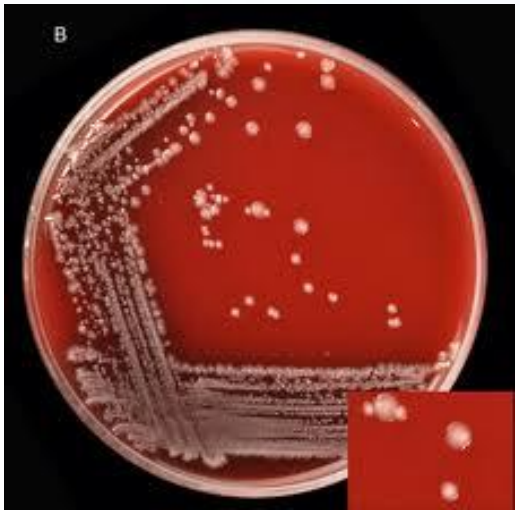
Chronic urinary infection with multi-drug resistant organisms



Accelerating microbiology

Potential to revolutionise stewardship

Now….



24 h to pure culture

48 h to susceptibility data

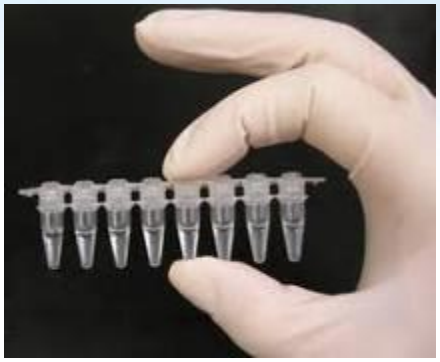
Meanwhile the patient is on empirical R_x
May be inappropriate --- or unnecessarily broad

Courtesy David Livermore

Accelerating microbiology

Potential to revolutionise stewardship & therapy

Future….



PCR on specimen
Recognise key pathogens and a few resistances

… some systems available

Next generation sequencing
Comprehensive pathogen and resistance detection

….under development

Potential to deliver results in 6h benefitting patient and stewardship…. but much work still to do

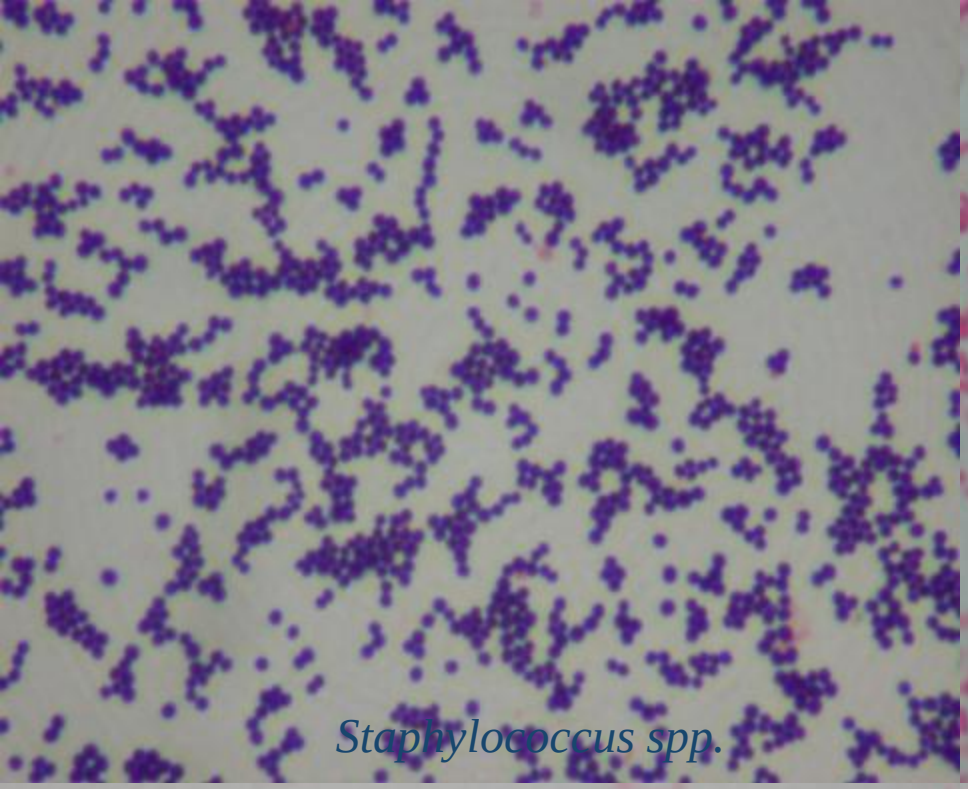
Courtesy David Livermore

What do we want rapid diagnostics to do?

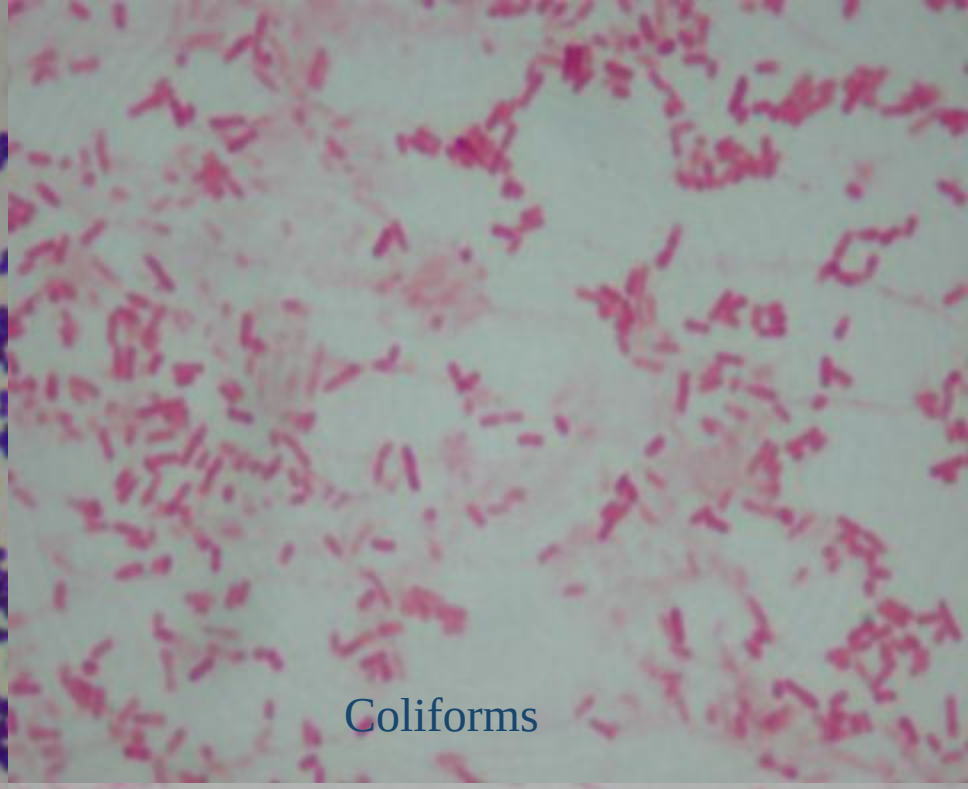
- Identify bacterial infection
- Support starting or withholding Abx
- Exclude bacterial infection
- Identify the pathogen
- Detect antibiotic resistance and guide Abx Rx
- Support stopping Abx
- Predicting prognosis

Identify infection

- **Rapid tests for specific germs**
  - **Influenza PCR**
  - **Malaria antigen detection**
  - **Group A/B strep detection**
- **What is wrong with a Gram stain?**



Staphylococcus spp.



Coliforms

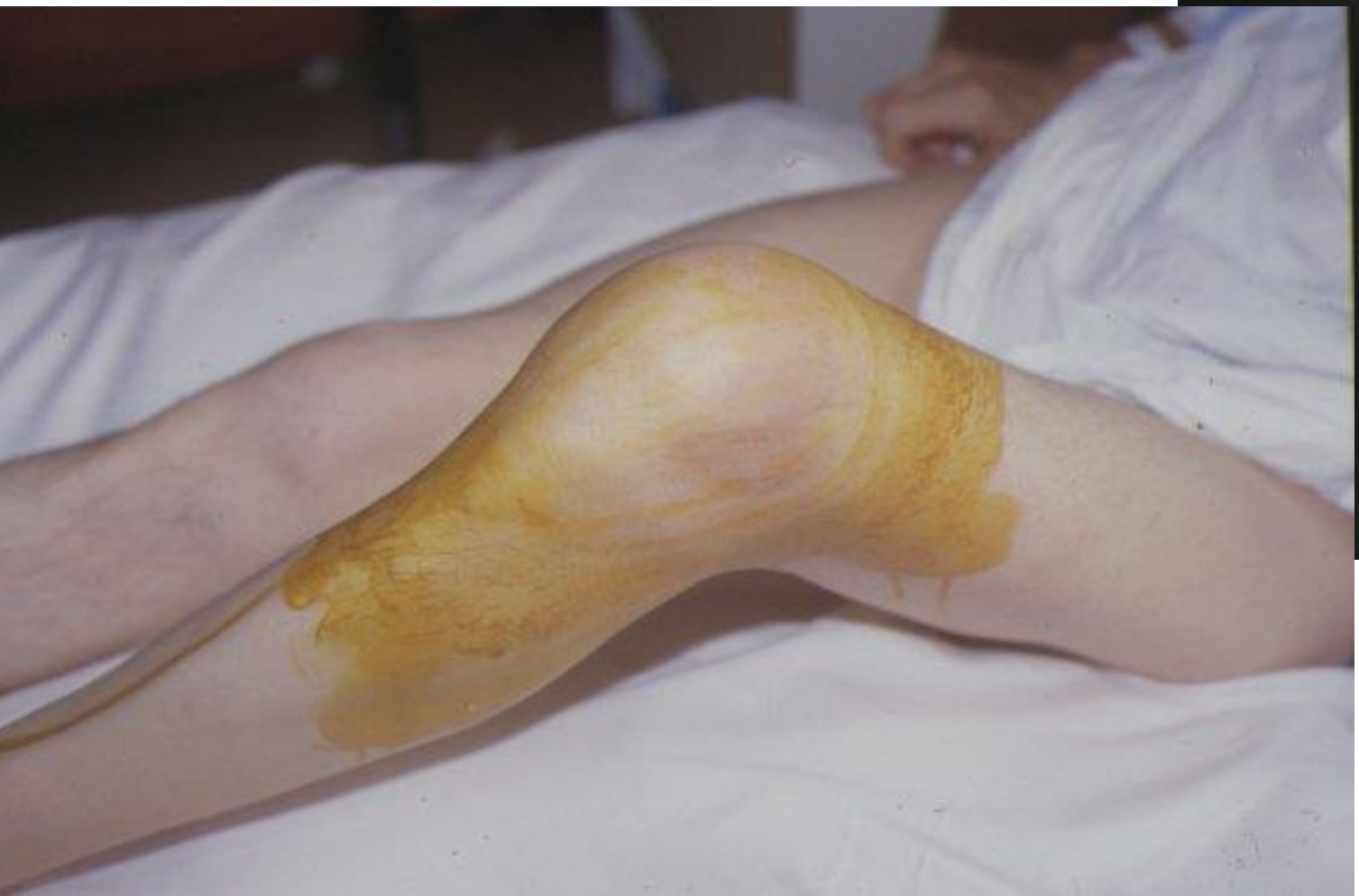
Fusiforms

Campylobacter spp.

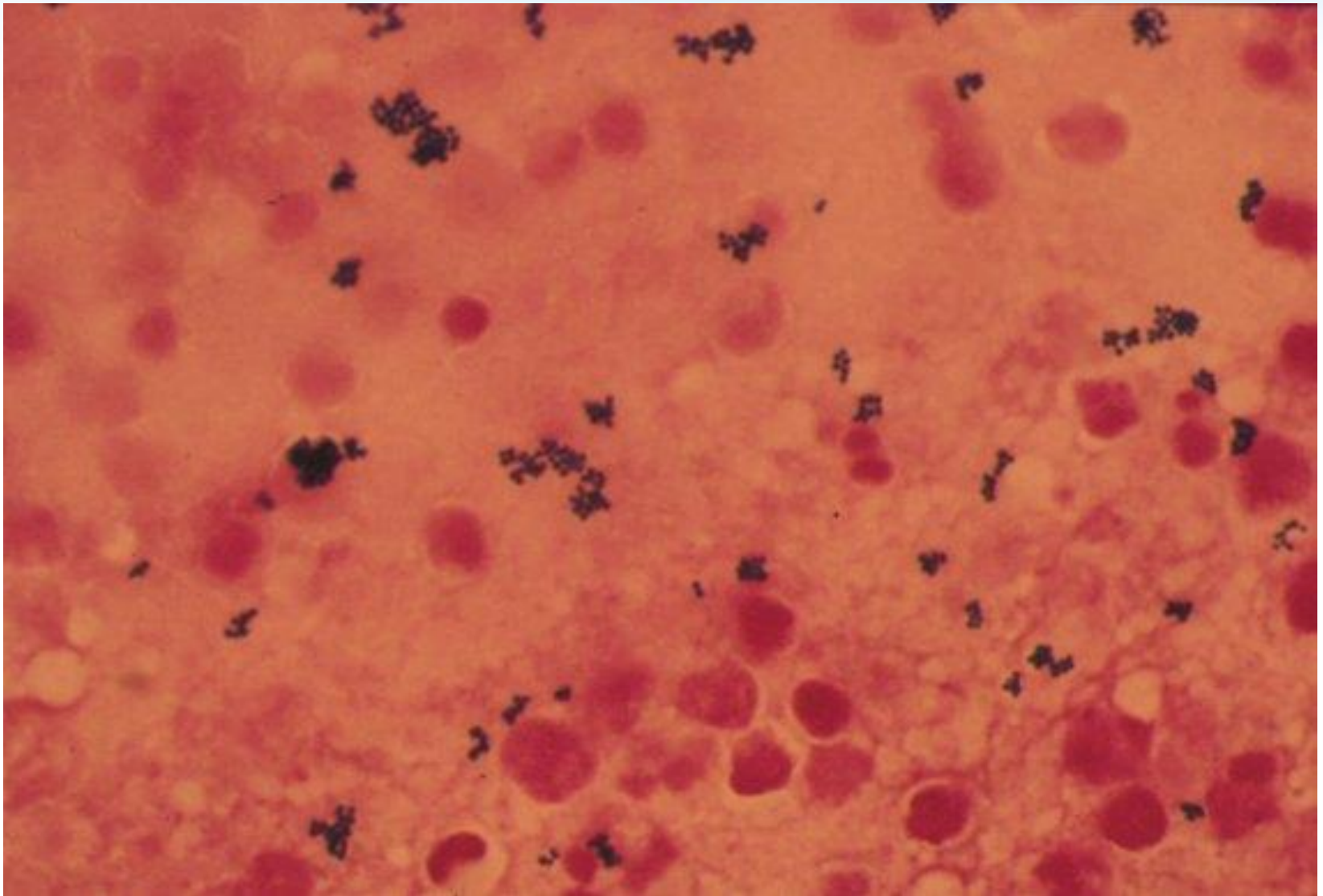
What Can a Gram Stain Tell You?

- **Gram Stain:**
  - **Presence or absence of WBC's - quantity**
  - **Presence or absence of epithelial cells - quantity**
  - **Presence or absence of bacteria - quantity**
  - **Presence or absence of yeast/fungal elements - quantity**
  - **Bacterial morphology - cocci, rods, coccobacilli**
  - **Gram reaction**
    - **Gram positive - blue**
    - **Gram negative - red**

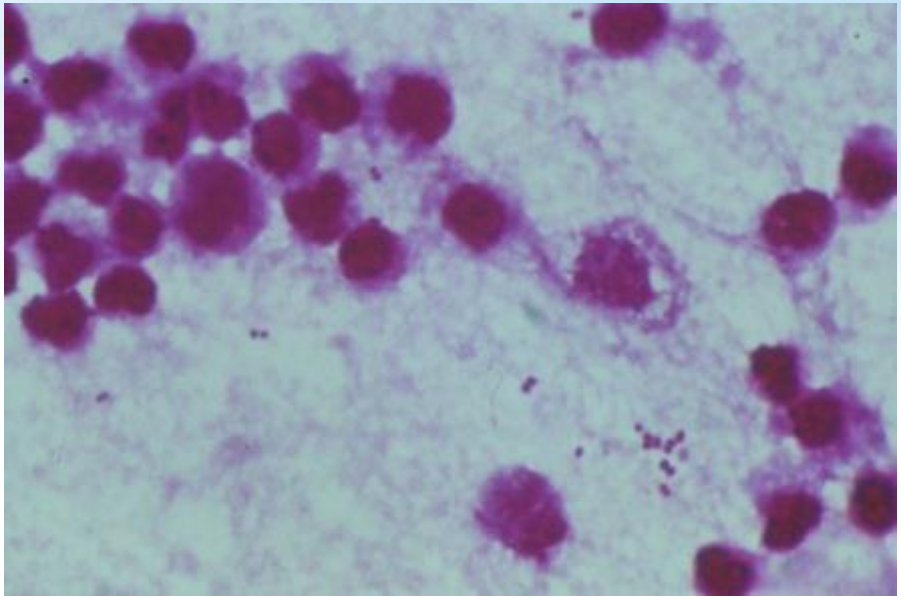
Septic arthritis

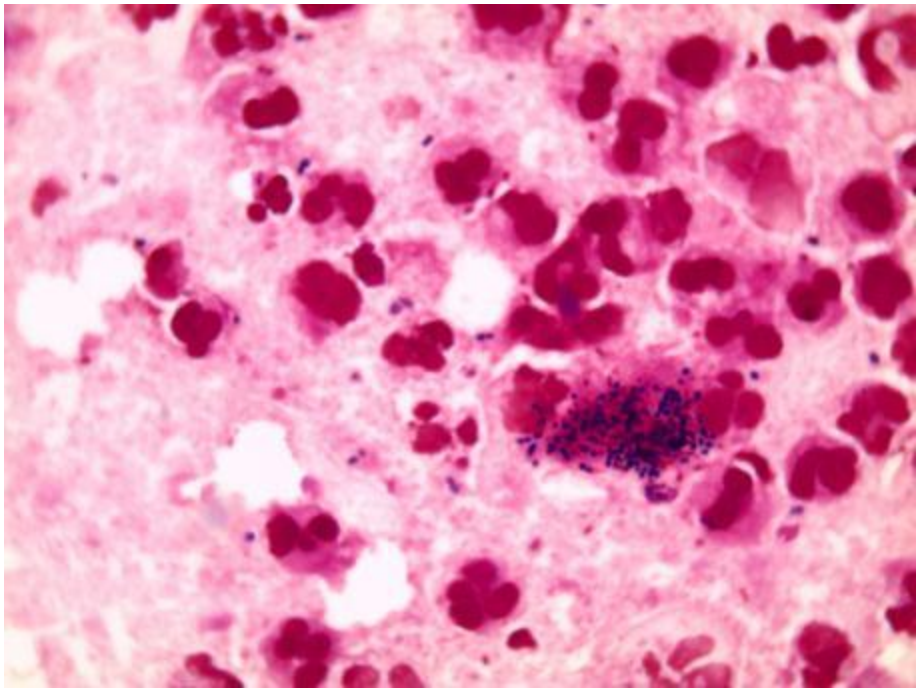


Knee joint pus



CSF - Grams





Support starting or withholding antibiotics

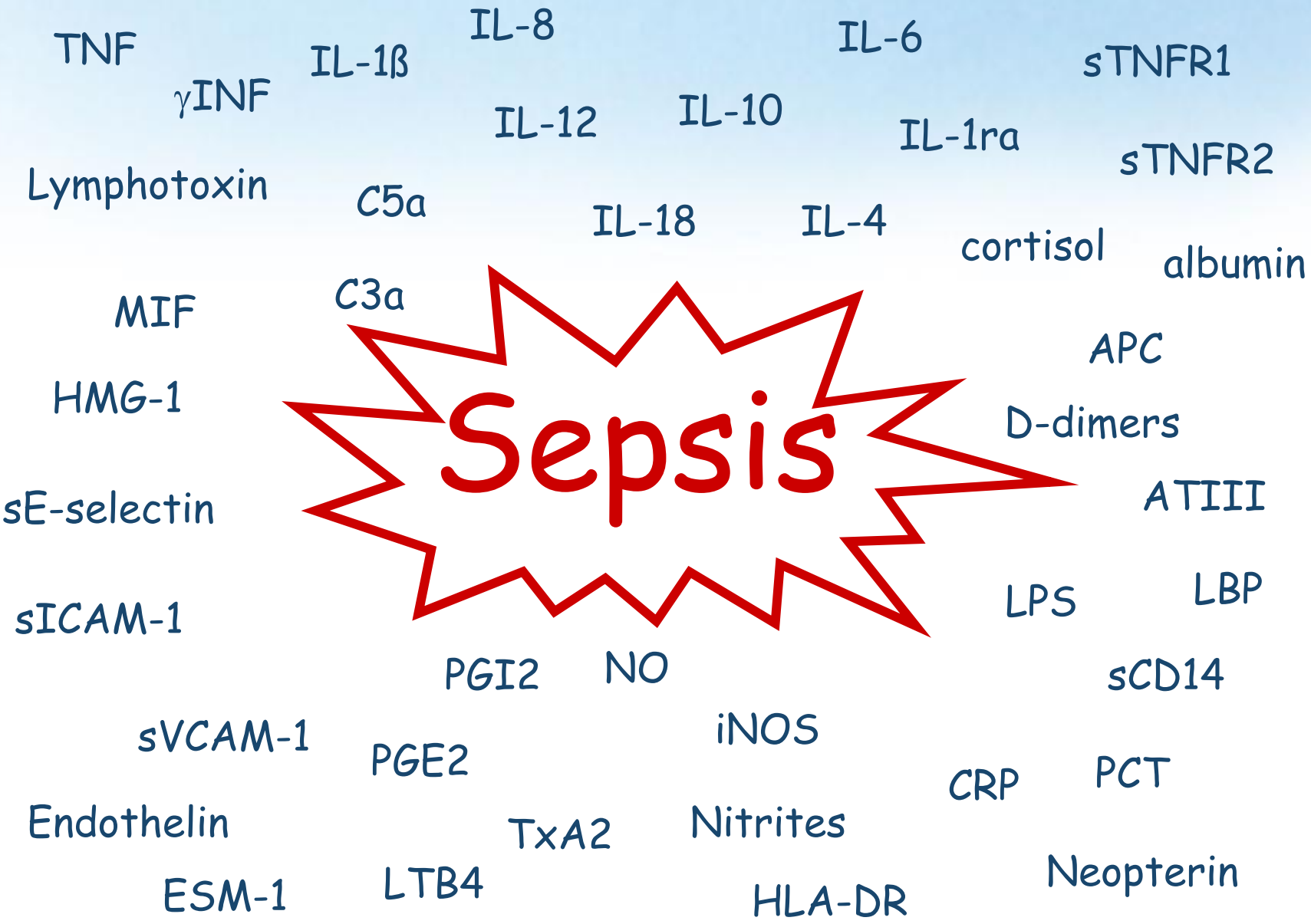
- Biomarkers
  - WBC
  - CRP
  - PCT

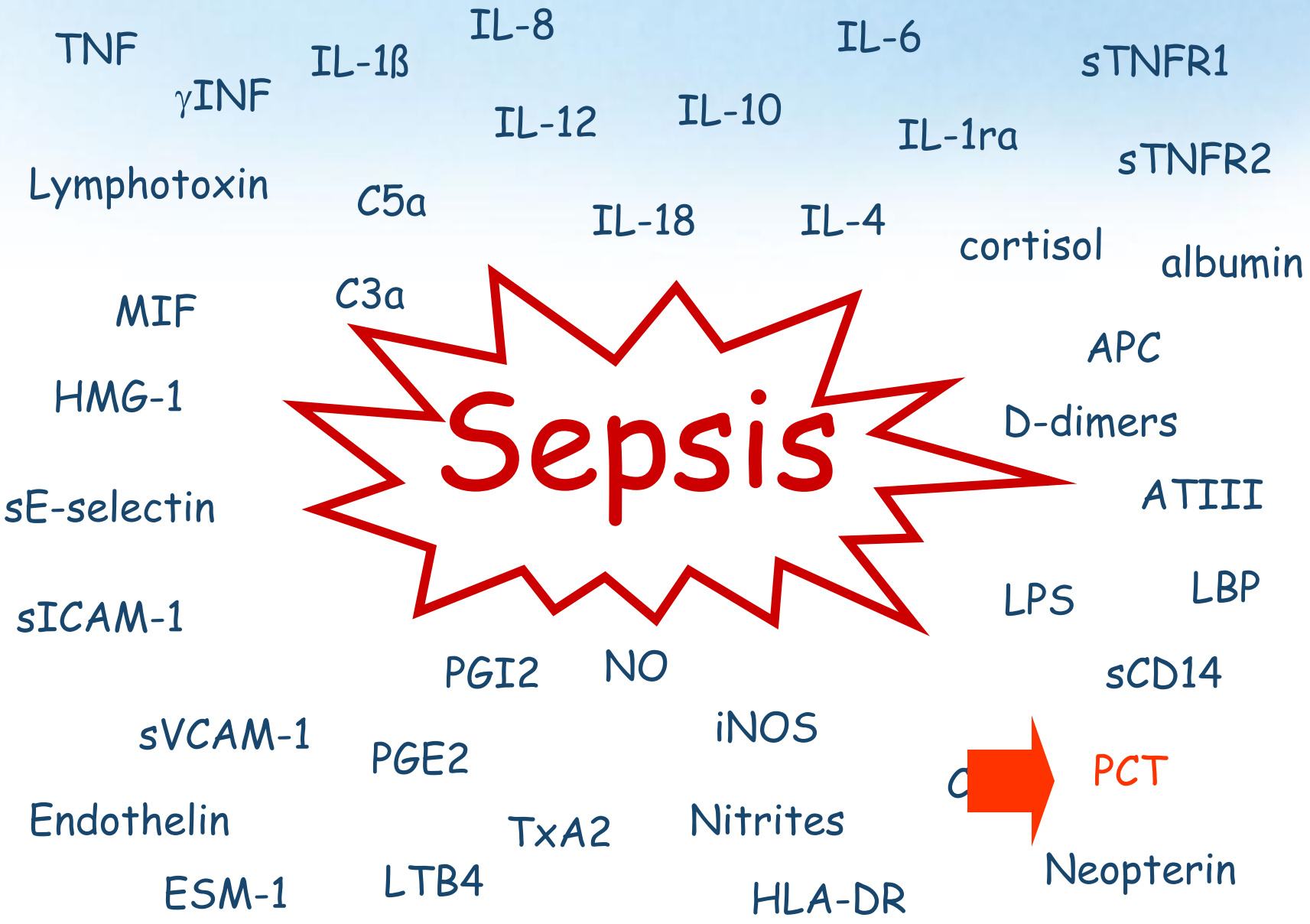
What was the reason you initiated the last prescription of antibiotic therapy?
Straw poll of F1, F2 doctors, ED dept

"Clinical Infection"
"Fever"
" Off legs"
" Mucky urine"
" ? aspiration"
" Increased respiratory requirements"
"Rising CRP"
"Wound infection"
"The boss asked me to"
"Positive culture"
…

Hampshire Hospitals Antibiotic week

Antibiotic user audit 2014


TNF
IL-1ß
IL-8
IL-6
sTNFR1
γINF
IL-12
IL-10
IL-1ra
sTNFR2
Lymphotoxin
C5a
IL-18
IL-4
cortisol
albumin
C3a
MIF
APC
HMG-1
Sepsis
D-dimers
sE-selectin
ATIII
LPS
LBP
sICAM-1
PGI2
NO
sCD14
sVCAM-1
iNOS
PGE2
CRP
PCT
Endothelin
TxA2
Nitrites
Neopterin
ESM-1
LTB4
HLA-DR


TNF
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Lymphotoxin
C5a
IL-18
IL-4
cortisol
albumin
C3a
MIF
APC
HMG-1
Sepsis
D-dimers
sE-selectin
ATIII
LPS
LBP
sICAM-1
PGI2
NO
sCD14
sVCAM-1
iNOS
PGE2
PCT
Endothelin
TxA2
Nitrites
Neopterin
ESM-1
LTB4
HLA-DR

Yafang Huang, Rui Chen, Tao Wu, Xiaoming Wei and Aimin Guo

Association between point-of-care CRP testing and antibiotic prescribing in respiratory tract infections:

a systematic review and meta-analysis of primary care studies

Abstract

Background

Most patients with respiratory tract infections (RTIs) are prescribed antibiotics in general practice. However, there is little evidence that antibiotics bring any value to the treatment of most RTIs. Point-of-care C-reactive protein testing may reduce antibiotic prescribing.

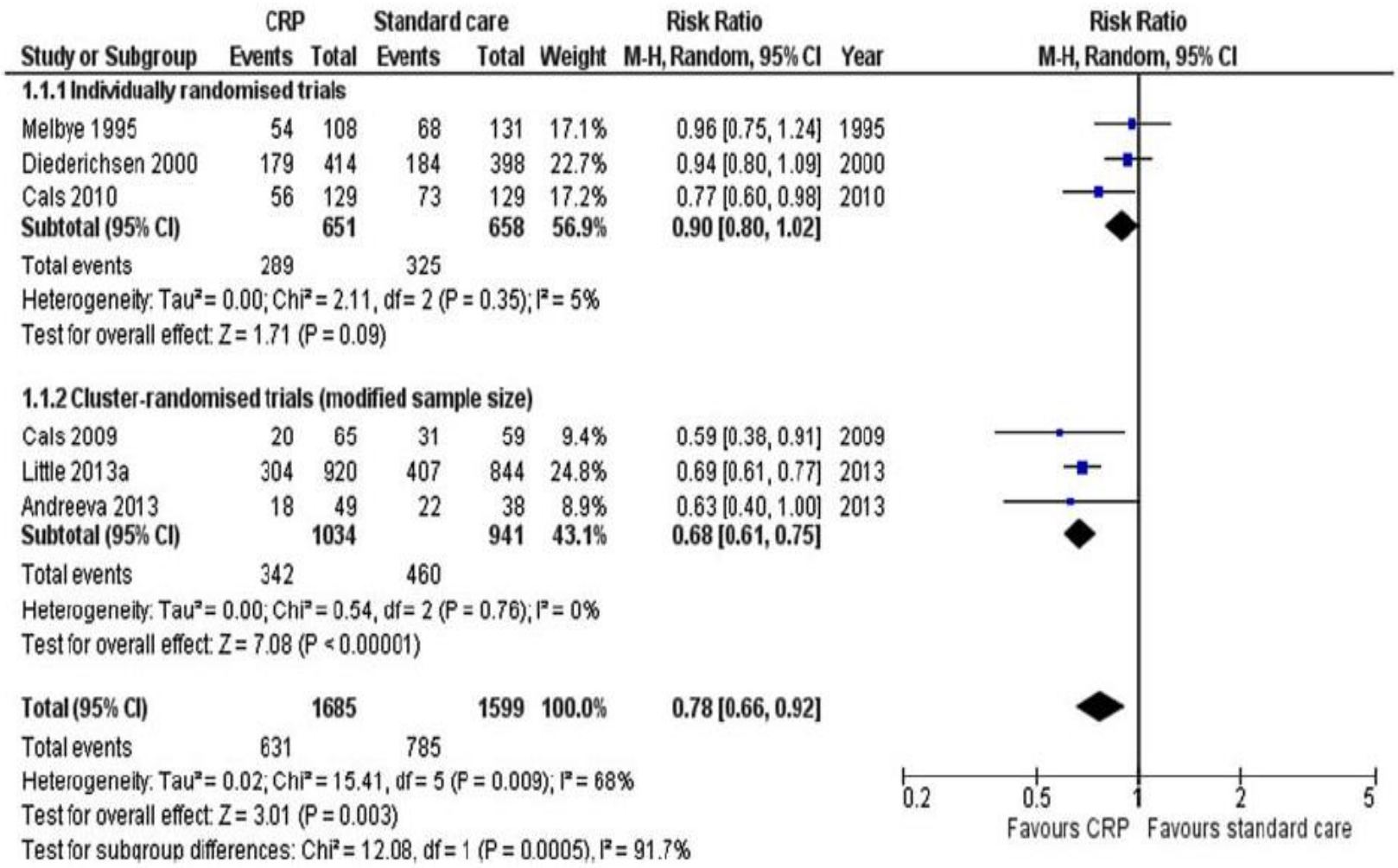
Aim

To systematically review studies that have

INTRODUCTION

Respiratory tract infections (RTIs) are among the most common acute conditions leading to patients seeking consultations in general practice.[1] About 80% of patients with RTIs are prescribed antibiotics.[2] However, RTIs are most often self-limiting and seldom require antibiotics for treatment.[3] The increased use of antibiotics is significantly associated with the development of drug-resistant bacteria. Clinical guidelines do

et al[13] concluded that POC CRP testing significantly reduced antibiotic prescribing for patients with RTIs. However, Gonzales *et al*[11] found there was no difference in antibiotic prescribing between POC CRP tested and control patients. In addition, these studies had small sample sizes and thus lacked a more convincing statistical power to clarify whether the use of POC CRP testing in general practice can reduce antibiotic prescribing.



Aabenhus R, Jensen Jens-Ulrik S, Jørgensen Karsten J, *et al.* Biomarkers as point-of-care tests to guide prescription of antibiotics in patients with acute respiratory infections in primary care. *Cochrane Database Syst Rev* 2014;11:CD010130. http://onlinelibrary.wiley.com/doi/10.1002/14651858.CD010130.pub2/abstract

Table 2 Criteria for management of RTIs in general practice with CRP POCT after proper clinical examination of the patient

Management	Draft NICE guidance for Pneumonia[49]	Dutch GP practice guideline[54]	GRACE study[14]	ERS[51]
Self-limiting RTI. Do not routinely offer antibiotic therapy. Pneumonia unlikely. Give education	CRP less than 20 mg/L	CRP less than 20 mg/L	CRP less than 20 mg/L	CRP less than 20 mg/L
Majority of patients have self-limiting LRTI. Assessment of signs, symptoms, risk factors and CRP is important. Withhold antibiotics, in most cases			CRP 21–50 mg/L	
Assessment of signs, symptoms, risk factors and CRP is crucial. Withhold antibiotics in the majority of cases and consider delayed antibiotics in the minority of cases			CRP 51–99 mg/L	
Consider a delayed antibiotic prescription. Clinical presentation decisive. Prescribe antibiotics only in patients with a high risk of complications*	CRP between 20 and 100 mg/L	CRP between 20 and 100 mg/L		
Severe infection. High risk of pneumonia. Offer antibiotic therapy	CRP greater than 100 mg/L	CRP greater than 100 mg/L	CRP greater than 100 mg/L	CRP greater than 100 mg/L

Cooke J, Butler C, Hopstaken R, *et al. BMJ Open Resp Res* 2015;**2**:e000086. doi:10.1136/bmjresp-2015-000086

CRP ≤ 20 mg/l
▪ Self-limiting LRTI ▪ Withhold antibiotics
CRP 21-50 mg/l
▪ Majority of patients have self-limiting LRTI ▪ Assessment of signs, symptoms, risk factors and CRP is important ▪ Withhold antibiotics, in most cases
CRP 51-99 mg/l
▪ Assessment of signs, symptoms, risk factors and CRP is crucial ▪ Withhold antibiotics in the majority of cases and consider delayed antibiotics in the minority of cases.
CRP ≥ 100 mg/l
▪ Severe infection ▪ Prescribe antibiotics

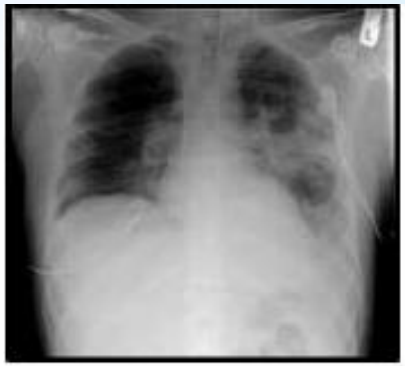
Little et al Lancet 2013

Procalcitonin study in patients with ‘possible’ infection

- 99 Medical Admissions Unit (MAU) patients with suspected infection
- 42 Intensive Care Unit (ICU) cases, with 87 procalcitonin tests done, suspected sepsis developing
- Half of antibiotic courses WITHELD as a result of LOW PCT
- Procalcitonin results delivered within 90 minutes of request
- Antibiotics withheld in 52/99 MAU cases and on 42/83 occasions in the ICU on basis of low procalcitonin
- No adverse effects from withholding antibiotics.
- Suggests scope for much better tailoring of antibiotic use

Saeed K, Dryden MS, Bourne S, Paget C, Proud A. *Journal of Hospital Infection* 2011;78(4),289–292;doi:10.1016/j.jhin.2011.03.018

Clinical pathways and PCT



B. Moderate and high acuity Patients

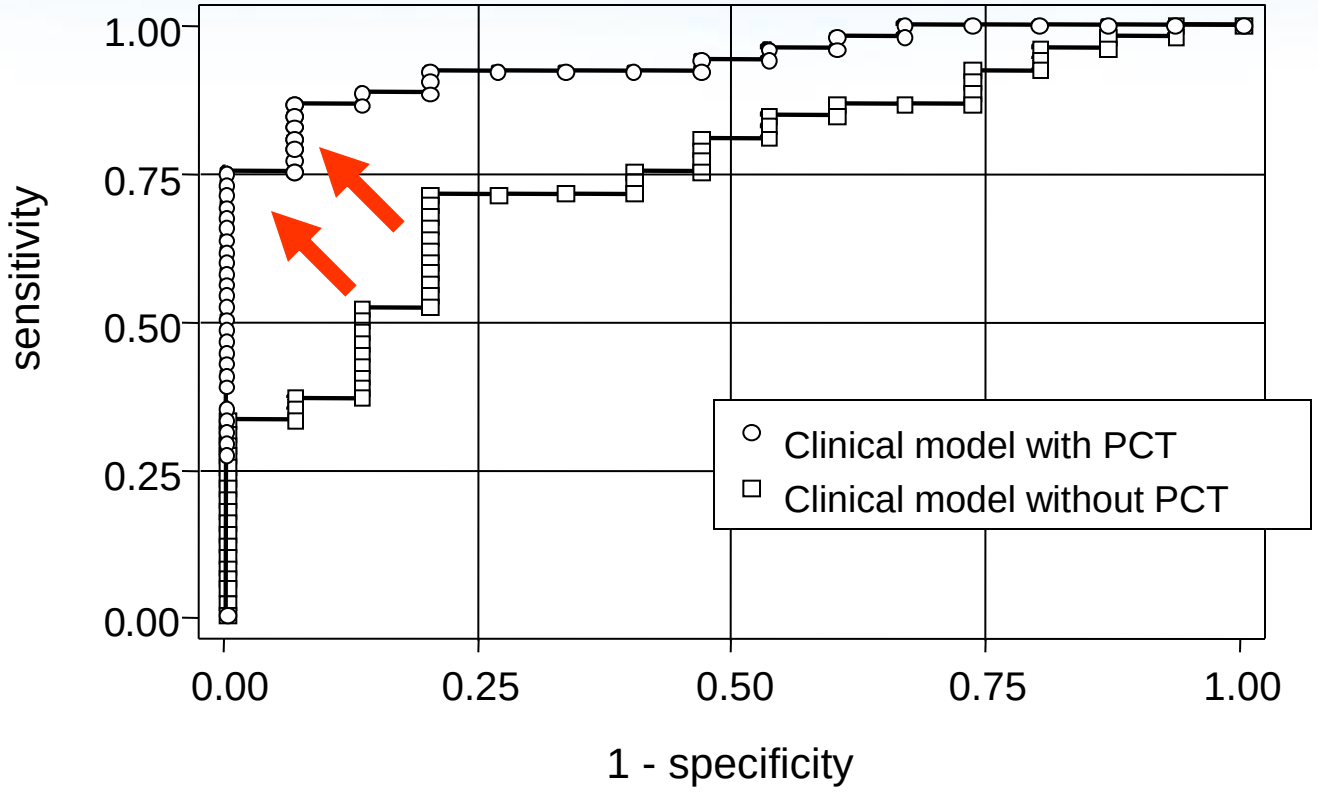
(CAP patients in ED, hospital ward or ICU setting)

Procalcitonin (µg/L)

	<0.1ug/L	<0.25ug/L	≥0.25ug/L	>0.5ug/L
Diagnosis	Bacterial infection highly unlikely; → consider alternative diagnosis	Bacterial infection unlikely → consider alternative diagnosis	Bacterial infection likely	Bacterial infection / sepsis highly likely
Prognosis	Low risk for mortality despite high clinical risk score	Low risk for sepsis related complication	High risk for bacteremic infection	High risk for bacteremic infection and adverse outcome -> monitor PCT for treatment response
Therapy	Consider AB treatment if high clinical suspicion of infection ("overruling") -> monitor PCT for early stopping AB treatment	Consider AB treatment if high clinical suspicion of infection ("overruling") -> monitor PCT for early stopping AB treatment	Start AB -> monitor PCT for stopping AB treatment if decrease >80-90% or PCT <0.25ug/L (ward) or <0.5ug/L (ICU)	Start AB -> monitor PCT for stopping AB treatment if decrease >80-90% or PCT <0.25ug/L (ward) or <0.5ug/L (ICU)

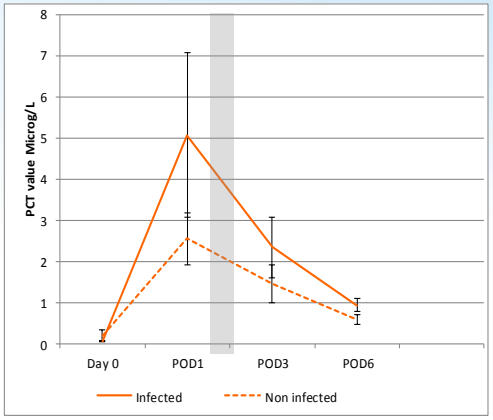
Schuetz P. et al Chest 2011

PCT adds to a clinical prediction model for the diagnosis of sepsis



Harbarth et al. Am J Respir Crit Care Med 2001

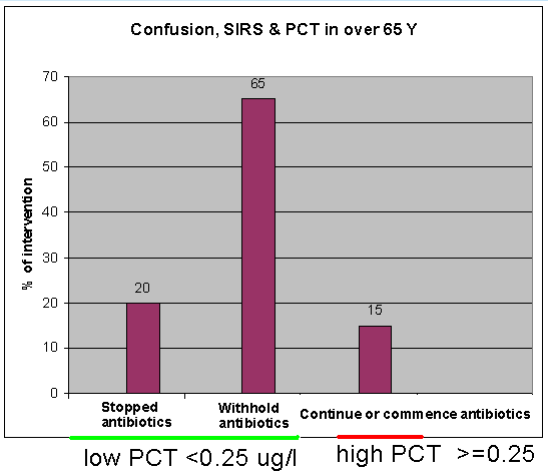
PCT HHFT summary



Pseudomyxoma

PCT predicts infections in pseudomyxoma patients
24hr prior to clinical suspicion (Gray line) and 48 hr prior to CRP and WCC (Data not shown)

Saeed K et al. *EJSO* in press, Oct 2015



Confusion and elderly ? UTI to avoid just in case Abx!

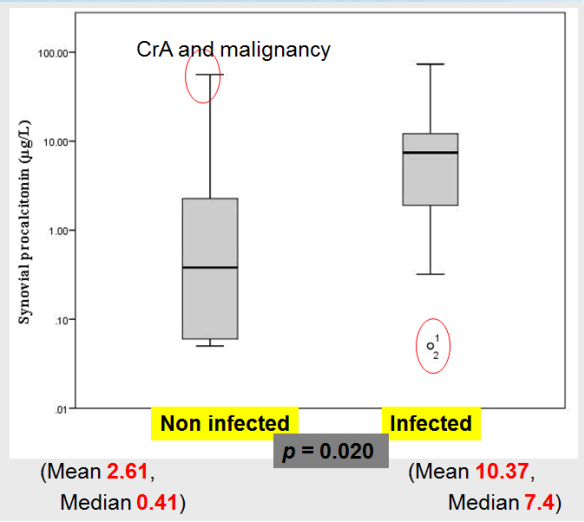
55 patients (1 ward → 3 months)

Confusion ? + potential (chest, or urine or etc infection)

In low PCT group 4/ 46 patients RIP (> 14 days after the PCT decision non due to infection)

In High PCT group 6/9 patients RIP all on antibiotics deemed appropriate

Eddie F, et al. *J Infect Non Infect Dis* 2015



SYNOVIAL PCT

Synovial PCT higher in SA vs non SA (caution with crystal arthropathy and concomitant malignancy

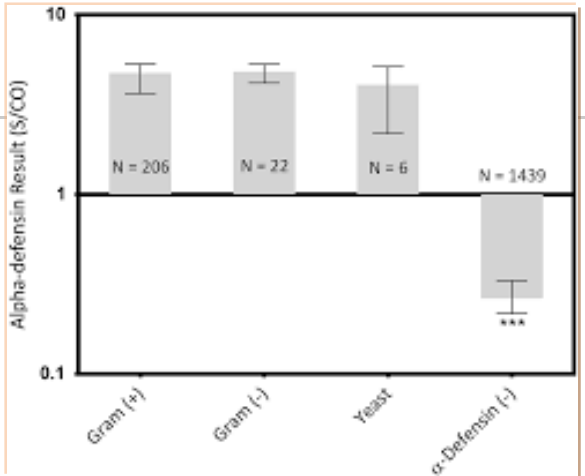
Synovial fluid PCT <0.5 µg/L → NPV of 0.90
More promising in excluding PJI

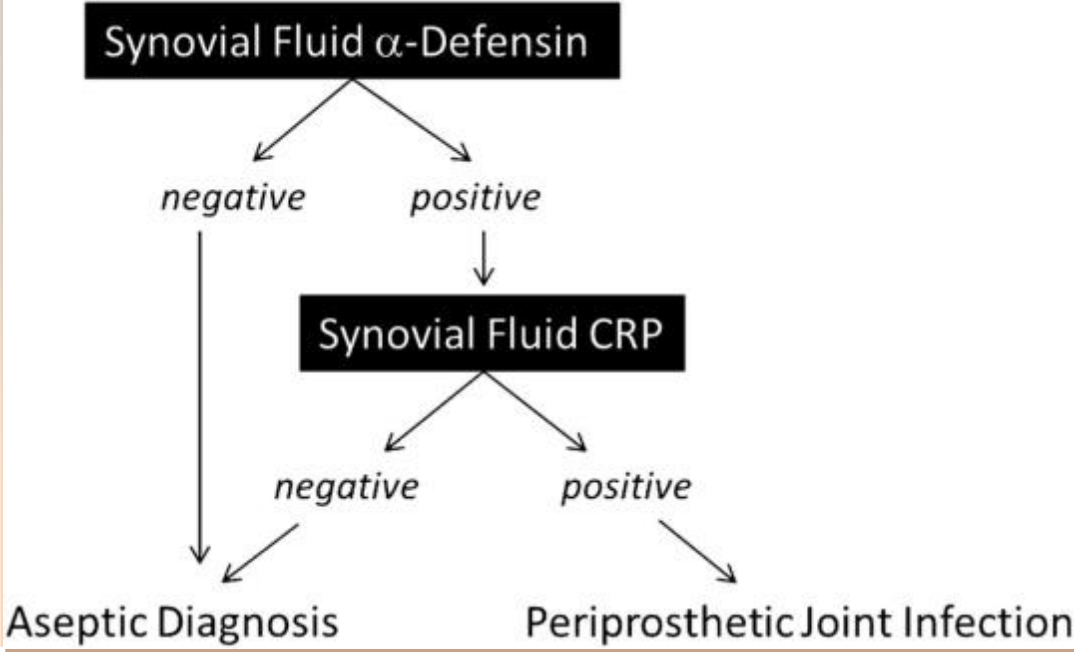
Saeed K, Dryden M et al. Infection. 2013; 28: 201-206

Courtesy Kordo Saeed

Exclude bacterial infection

- The *Synovasure* Test achieves 97% sensitivity and 96% specificity by measuring synovial fluid alpha defensin
- Alpha defensin is an antimicrobial peptide released by neutrophils in response to pathogens
- Alpha defensin is an ideal biomarker for PJI due to the tremendous separation it achieves between positives and negatives





Synovasure Performance		95% Confidence Interval
Sensitivity	97.4%	86.1% - 99.6%
Specificity	95.8%	90.5% - 98.6%

Cognitor Minus (Momentum Bioscience Ltd)

- **A CE marked product to confirm absence of bloodstream infection in routine blood cultures the day after specimen receipt by lab**
- **Uses proprietary ETGA technology to provide universal detection of viable bacteria and fungi via qPCR**
  - **Does not identify organism**
  - **Not designed for confirming positive infection**
- **Provides 99.5% negative predictive value at 1 day *vs* blood culture after 5 days**
- **Clinical data indicate Cognitor Minus also detects positive specimens missed by conventional culture**

Cognitor Minus implementation

- Uses existing lab equipment including qPCR machine
  - Only additional equipment is ‘bead beater’ supplied FOC.
- Uses existing blood culture specimens
  - One test uses 0.5mL from each blood culture bottle in set
  - Bottles returned to incubator for continued analysis
  - Does not interfere with normal operation of culture bottles
- Test run on bottles negative after >12hr in automated blood culture cabinet
- Designed to encourage early cessation of antibiotics, reduce antibiotic-associated disease and reduce healthcare costs

Negative predictive value comparison

- **Septifast (Roche)*** **~90%**
- **Sepsitest™ (Molzym)**** **80-97%**
- **Iridica (Abbott)**** **97-99%**
- **Cognitor® Minus (Momentum Bioscience)***** **99.5%**

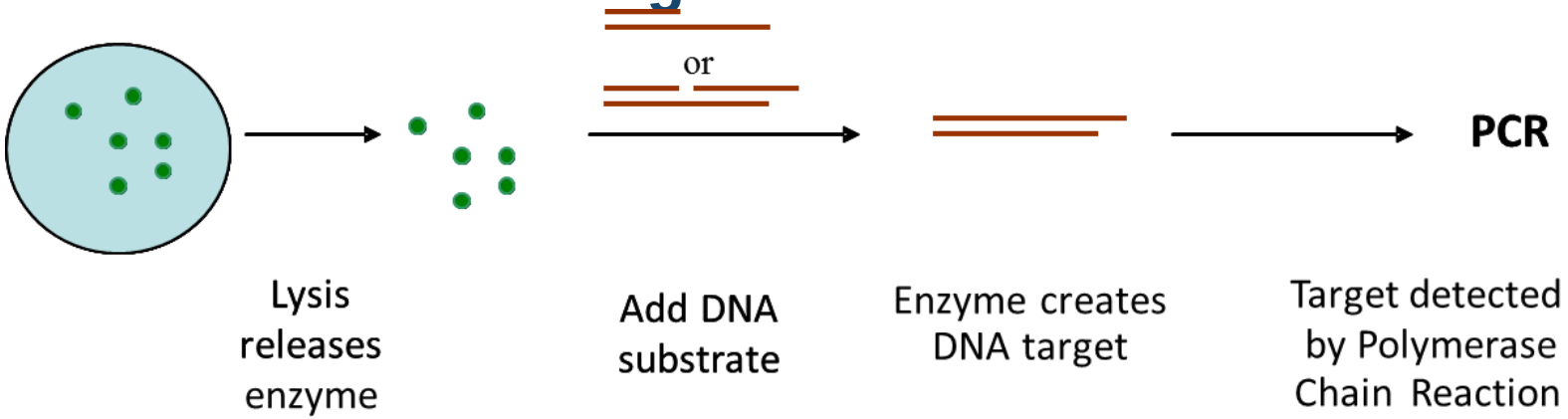
*** estimate based on prevalence of organisms detected by product**

**** data from published papers**

**** data from clinical performance evaluation on 1497 blood culture sets conducted at Hampshire Hospital Foundation Trust, December 2014 – March 2015**

Technology

- ETGA (Enzymatic Template Generation & Amplification) technology sensitively detects enzymes within micro-organisms
- Enzyme activity coupled to PCR-based DNA detection
- Unique combination of speed and universal detection of viable organisms



Features & Benefits

Unique rapid technology delivering:

- Improved result time for positive specimens
- Aids patient management and differential diagnosis
- Reduction in antibiotic use
- Detection of only viable organisms

Comparison with existing technology

Benefit	Culture	Molecular	ETGA
Rapid result		√	√
Only detects viable organisms	√		√
Detects all cultivable species	√		√
Detects non-cultivable organisms		√	√
Phenotypic AST	√		√

Clinical areas where Cognitor® Minus is likely to offer the greatest benefit

- Low risk of infection / dubious differential diagnosis
- Improving clinical picture but continuation of antibiotics is common
- Patient more susceptible to adverse effects of antibiotic treatment
- Confounding sign or symptom mimicking clinical picture.
- High cost of treatment, especially anti-fungal

- Clinical areas:
  - Oncology (solid tumours)
  - Emergency admissions
  - ICU
  - Neonates

✓Clinical role of Cognitor minus:

- ✓ Cessation of unnecessary Abx
- ✓ Deescalation of Abx
- ✓ Diagnostic algorithm
- ✓ Support IPC
- ✓ Reduce costs of Abx and antifungals

Momentum Cognitor

Cognitor Minus

- **Next-day detection of negative blood cultures**
  - Allows reporting of negatives 4 days earlier than currently
  - 99.9% NPV in trials on clinical specimens
  - Assists differential diagnosis and/or step down in care
  - Product ready to enter 6 month verification and validation phase prior to CE marking

Cognitor Plus

- **Same-day detection of positive blood cultures**
  - 92% sensitive, 99% specific in trials on clinical specimens
  - Product close to ready to enter verification and validation

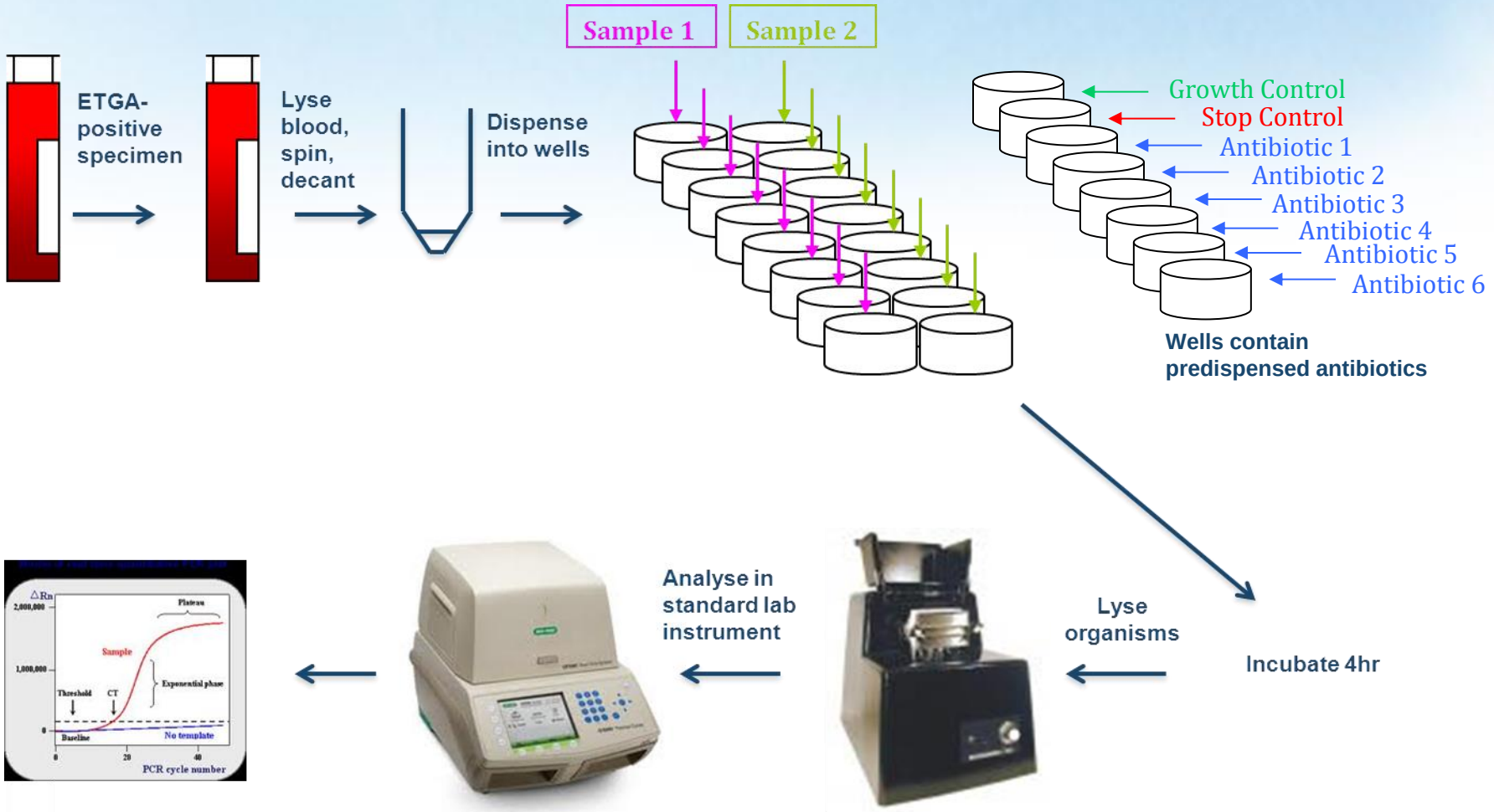
Momentum Cognitor ETGA system

Product 3

- **Detection and differentiation of Gram +ve/Gram -ve/fungi**
  - Still conceptual, based on response to antimicrobials
  - May also produce a stand-alone fungal detection test using either
    - Ligase detection of eukaryotic microorganisms
    - Antibiotic-loaded pre-culture

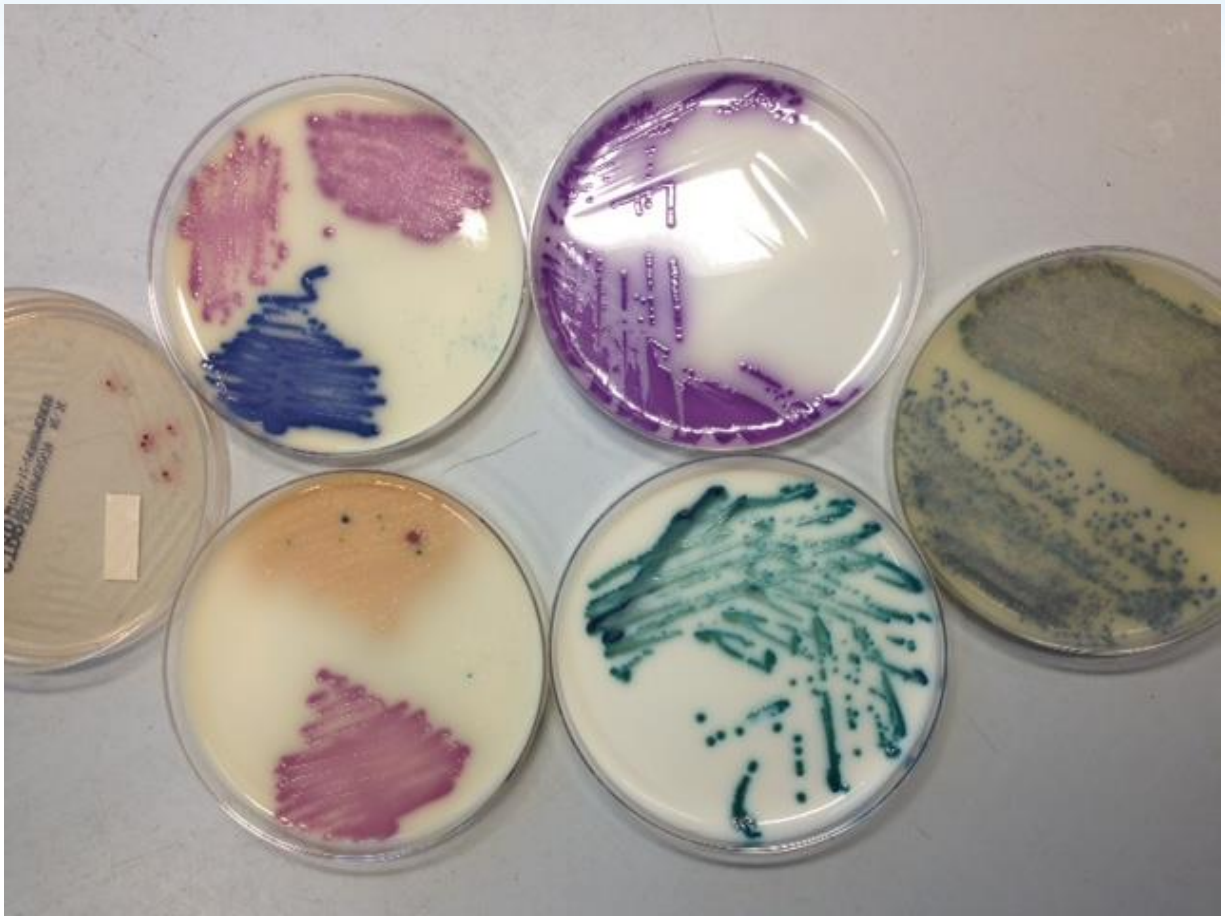
Product 4

- **24hr antibiotic susceptibility test**
  - Results within 24 hours of sample reaching the lab
  - Detection in presence of antimicrobials provides phenotypic result
  - Company has been awarded £88k Phase 1 SBRI contract to show proof of feasibility and expects to apply for £1-2m Phase 2 award in 2013



ETGA-AST concept

Identify the pathogen



Standard Microbiology

Media –

→ Selective

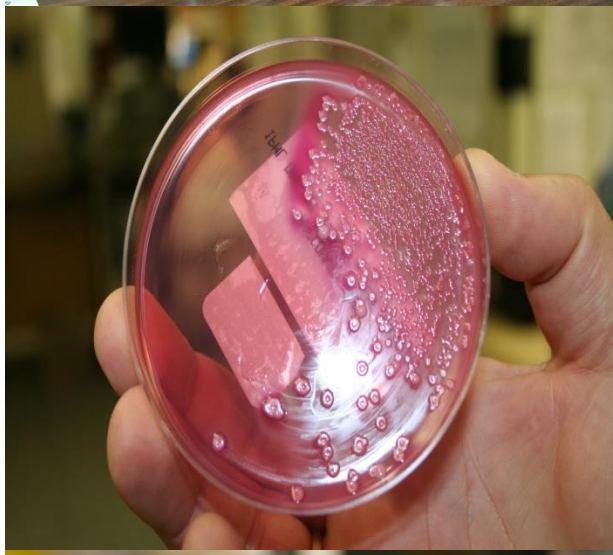
- selective growth of particular organisms,
  i.e. TSA and SRBC and antibiotics.
  Thayer-Martin/GC
  MacConkey

→ Differential

- for the differentiation of organisms,
  i.e. Sorbitol MacConkey; XLD, TCBS

→Chromogenic for resistance and screening

- → For MRSA
- → VRE
- → Candida ID
- → Gp B strep screening
- → ESBL, CPE



The Future of Clinical Microbiology

- **Less traditional microbiology**
  - **Flow cytometry**
    - **Screening urine for presence of bacteria prior to culture – human medicine ~35% NG – immediate reporting**
  - **Mass spectometry**
    - **Organism identification in ~5-30 minutes from bacterial colony or directly from specimen – i.e. fluid specimen (aspirate) of blood culture**
    - **Some issues with identification of Streptococci**

Multiplex PCR and ETGA

Rapid ID of selection of pathogens and some resistance mechanisms

- **Septifast – 25 organisms from whole blood**
- **Sepitest – many microbes detected by sequencing**
- **Iridica**
- **Cognitor plus**
- **All in development and need further clinical validation**

1 Test. 27 Targets. All in About an Hour

Set up a Run in 2 Minutes & Results in About 1 Hour

During the ICAAC 2013 Annual Meeting, come see what everyone is talking about.

The FilmArray Blood Culture Identification (BCID) Panel tests for a comprehensive list of 24 pathogens and three antibiotic resistance genes associated with bloodstream infections. With just one test you can identify pathogens in 9 out of 10 positive blood cultures in about an hour with only two minutes of hands-on time.

Ask about our other panels

- Respiratory Panel
- Gastrointestinal Panel*
- Meningitis Panel*
- Lower Respiratory Panel*

* In development

Get faster results at **FilmArray.com**

Blood Culture Identification Panel

Pathogens

Gram-Positive Bacteria

Enterococcus
Listeria monocytogenes
Staphylococcus
Staphylococcus aureus
Streptococcus
Streptococcus agalactiae
Streptococcus pneumoniae
Streptococcus pyogenes

Gram-Negative Bacteria

Acinetobacter baumannii
Haemophilus influenzae
Neisseria meningitidis
Pseudomonas aeruginosa
Enterobacteriaceae
Enterobacter cloacae complex
Escherichia coli
Klebsiella oxytoca
Klebsiella pneumoniae
Proteus
Serratia marcescens

Yeast

Candida albicans
Candida glabrata
Candida krusei
Candida parapsilosis
Candida tropicalis

Antibiotic Resistance Genes

mecA - methicillin resistance
vanA/B - vancomycin resistance
KPC - carbapenem resistance

BIOFIRE Idaho Technology is now BioFire Diagnostics, Inc.

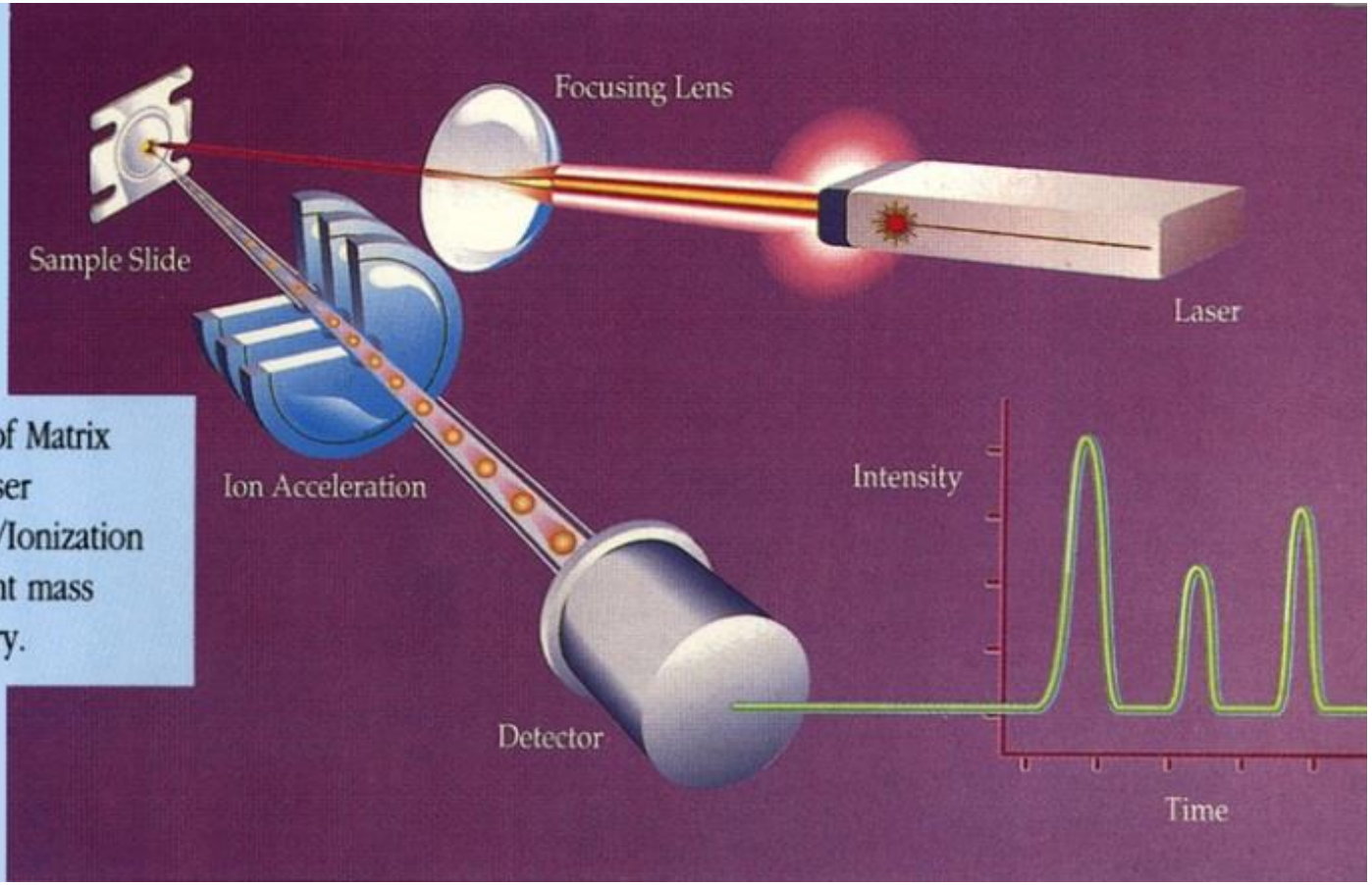
MALDI-TOF MS: How does it work?

- Start with a single bacterial colony

- Put on a target slide
- Quality control well inoculated with ATCC *E. coli*
- Addition of a matrix

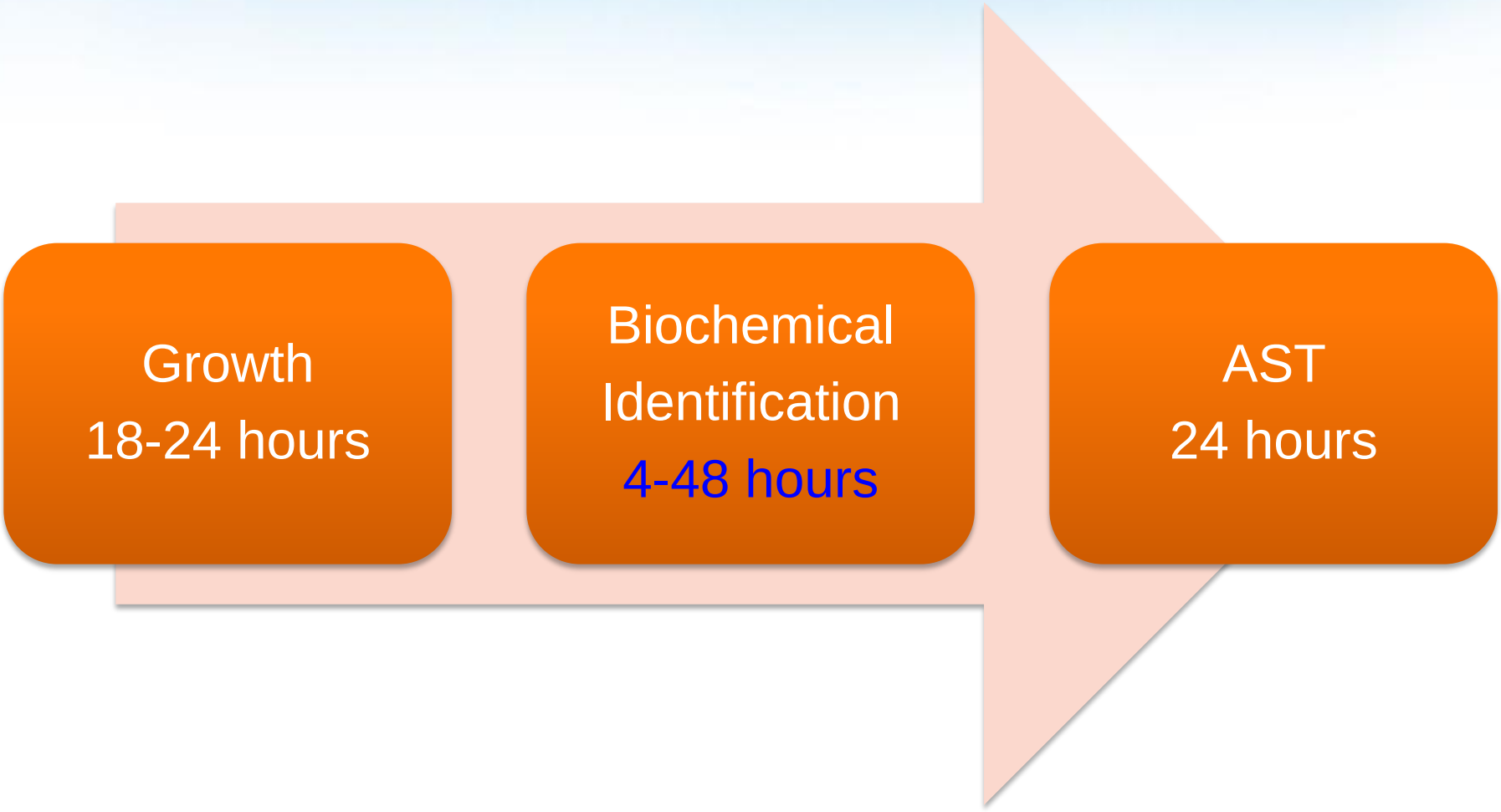
  1 ul of α-cyano-4-hydroxycinnamic acid in 50% acetonitrile and 2.5% trifluoroacetic acid
- Addition of formic acid if yeast

MALDI-TOF MS: How does it work?

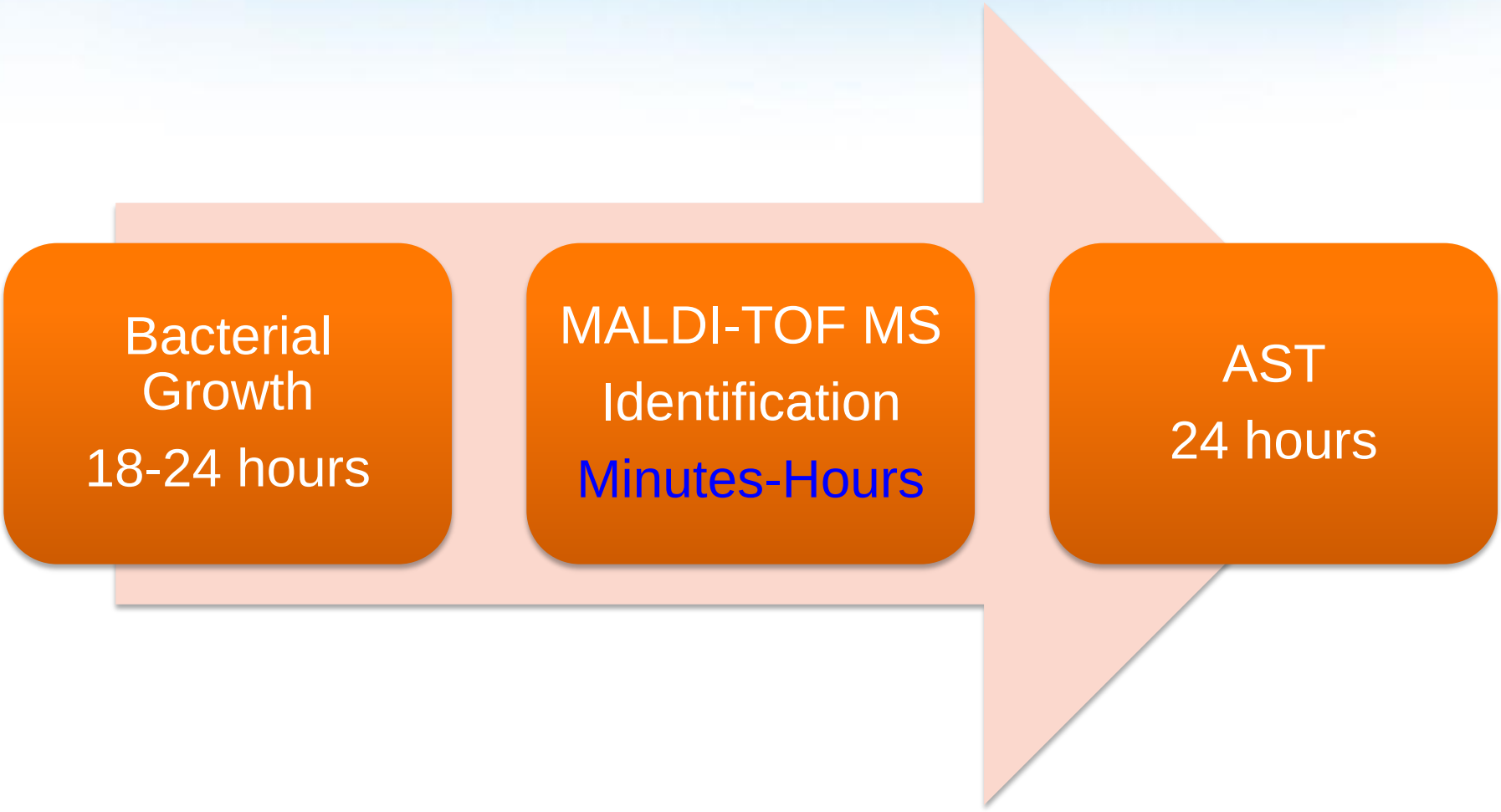


Schematic of Matrix Assisted Laser Desorption/Ionization time-of-flight mass spectrometry.

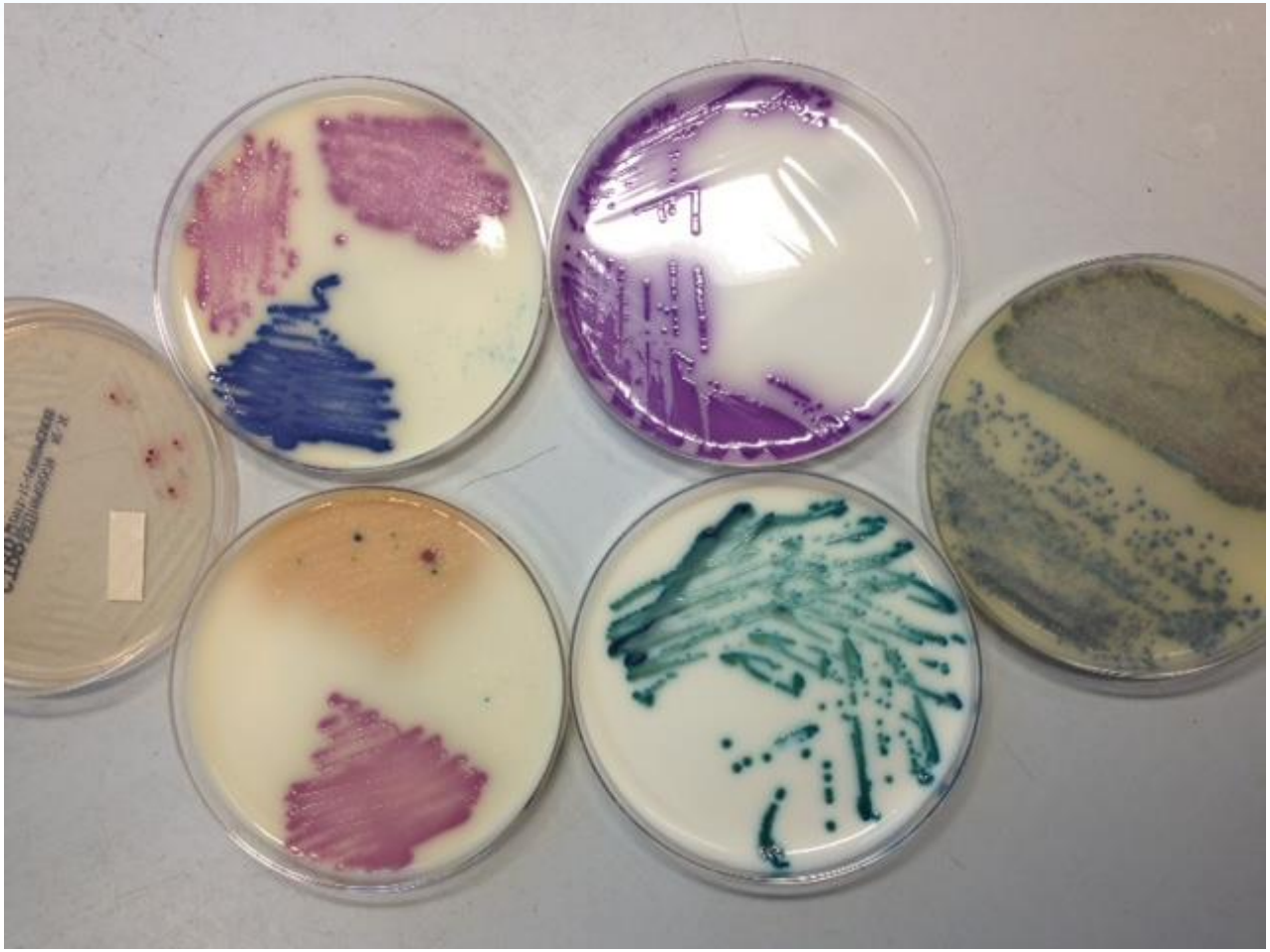
Laboratory Implementation: Before



Laboratory Implementation: After



Detect antibiotic resistance



The Superbug Challenge

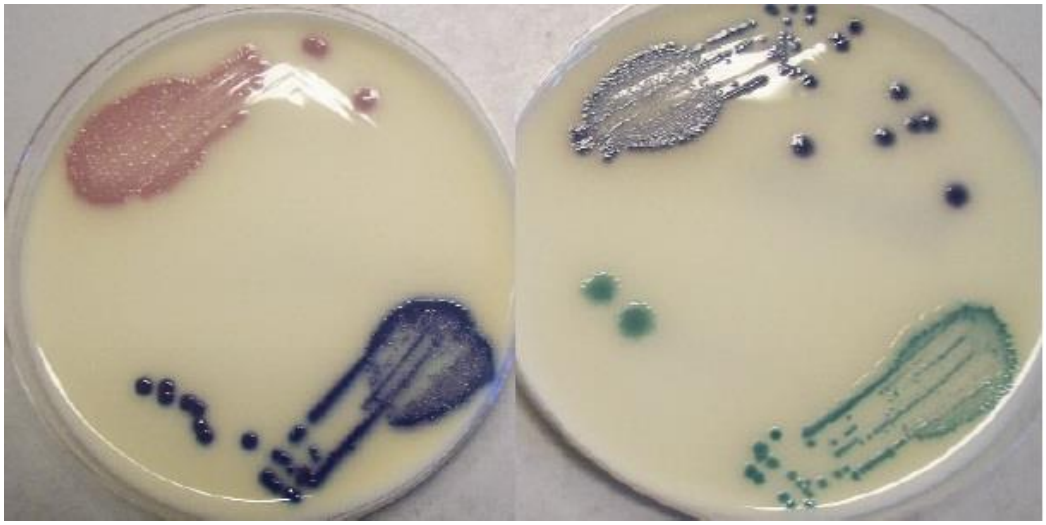
Acronym	Definition	Screening	Bacteria	Significance
ESBL	Extended spectrum	R to 3rd gen cephalosporins*	*E. coli*, *Kleb*. Spp. *Enterobacteriaceae*	R to most beta-lactams & cephalosporins
CPE		Carbapenems		
MRSA	methicillin R *S. aureus*	R to oxacillin PCR – *mec* A Chromo agar Cefoxitin R	*S. aureus*	R to all beta-lactams**
VRE	vancomycin R enterococcus	Van screen plate PCR-van genes chromo agar	*Enterococcus* spp.	R to vancomycin
VISA	Vancomycin inter	reduced S to vanco	*S. aureus*	reduced S to vanco
VRSA	Vancomycin R	resistance to vanco	*S. aureus*	R to vancomycin

*cefotaxime, cefpodixime, ceftriaxone, ceftazidime
** penicillins, cephalosporins, carbapenems, monobactams

Blondeau, JM, 2013, STAT – Steps to Antimicrobial Therapy, Companion Animals, 2nd Edition In Press: North American Compendium

Detecting AMR

- Chromogenics
- PCR
- Probes
- Cognitor ETGA



Support stopping antibiotics

- **Biomarkers**
  - **PCT**
  - **CRP**
- **Molecular**
  - **Cognitor minus**

Duration of antibiotic therapy in the ICU and elsewhere. Who cares?

Bacterial resistance

Toxicity - interactions

£££

Why did you stop the antibiotic therapy? Views form ICU

"We said 8 days"
" Micro round said stop"
"Patient is stable"
"Patient is transferring to the ward"
"Patient developed a rash"
"Renal function is deteriorating"
"New consultant on"
"Cultures came back negative"
"The PCT has fallen"
…

Hampshire Hospitals Antibiotic week

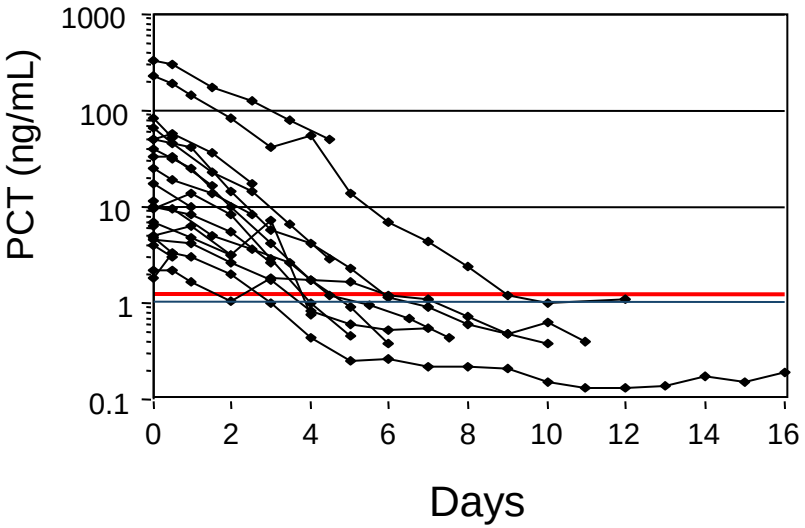
Antibiotic user audit 2014

Predicting Prognosis

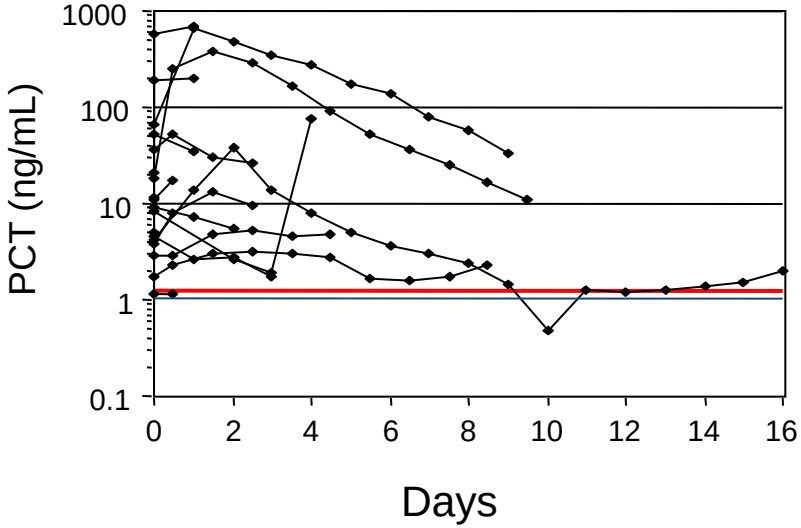
Prognostic tool?

The evolution of PCT levels with time predicts outcome

Survived

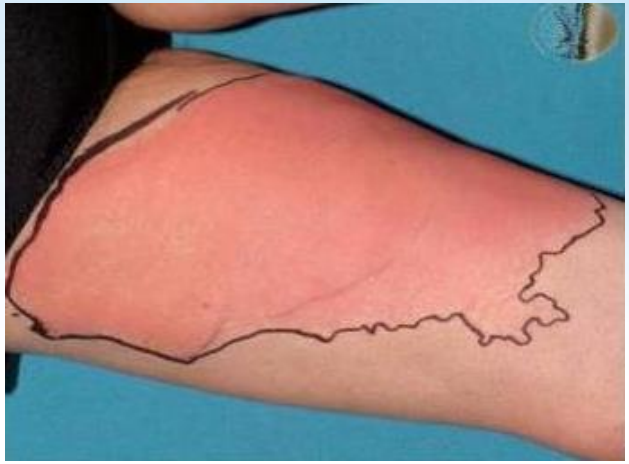


Died

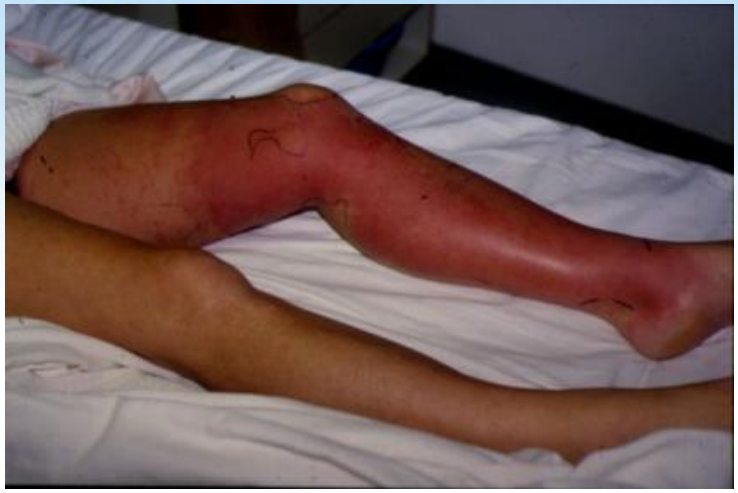


Would PCT help clinicians to stop antibiotics earlier ?

Harbarth et al. Am J Respir Crit Care Med 2001

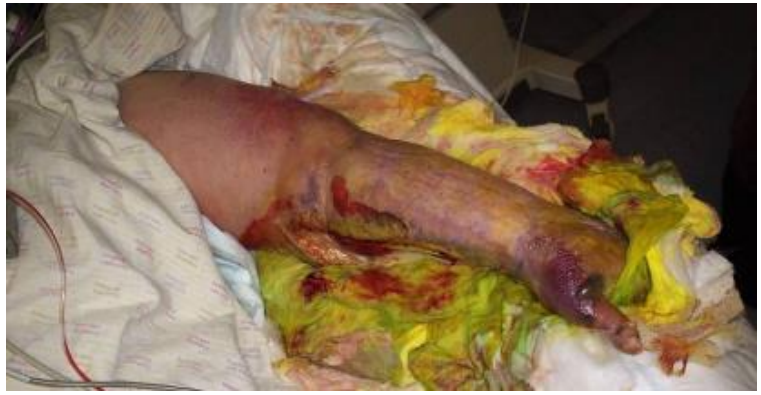


PCT = 0.75ng/ml



PCT = 1.8ng/ml – BC positive

- **Gp A streptococcal soft tissue infection**
- **A range of severity**



PCT = 26.2ng/ml – nec fasciitis

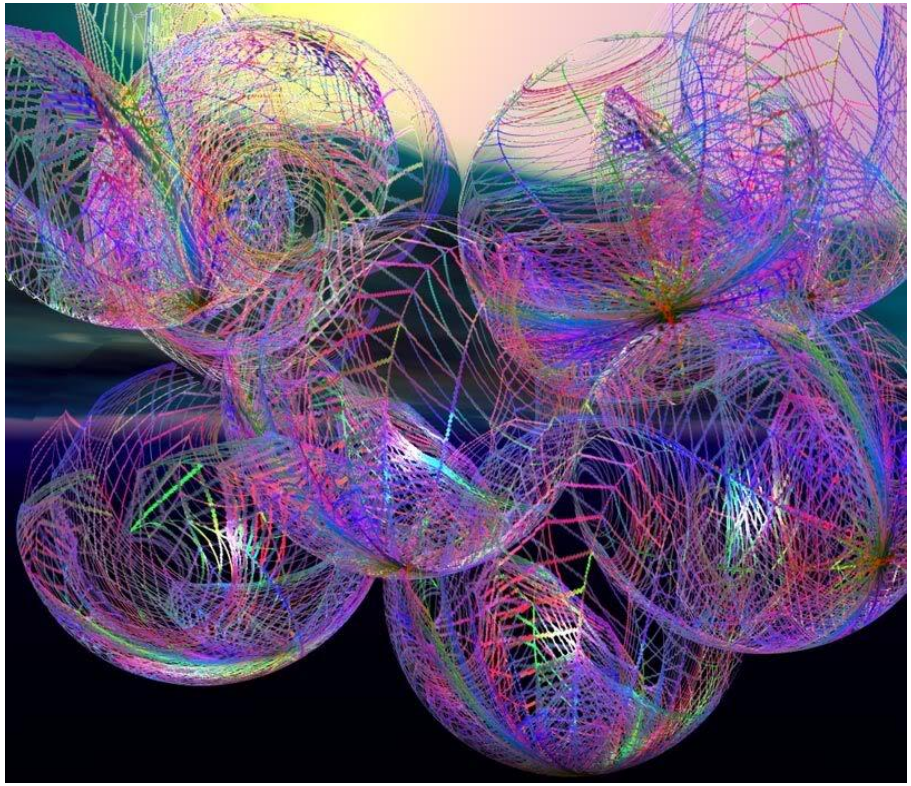
Summary

Identifying the pathogen	in situ or once cultured: MALDI-TOF, PCR, multiplex PCR, Nanoparticle Probe Technology , and PNA FISH, automated optical systems such as VITEK
Antimicrobial susceptibility	PCR, multiplex PCR, Nanoparticle Probe Technology Automated real time PCR, VITEK, Cognitor technology ETGA
Presence of bacterial infection	Cognitor Momentum ETGA in BC
Absence of bacterial infection	Cognitor minus Momentum ETGA
Biomarkers to predict the presence of bacterial infection	PCT, CRP, pro-adrenomedullin, alpha-defensin
Prognosis and course of infection	CRP, PCT, pro-adrenomedullin
Diagnosis and disease stage	Metabolomics / proteomics is in its infancy for the detection of infection as opposed to the identification of cultured bacteria.

‘Think beyond the 11th dimension of hyperspace’

‘Run your finger round the rim of a black hole’

Today program, Radio 4, Nov/Dec 2015



Thank you