

# Wound Infection and Biofilm Treatment

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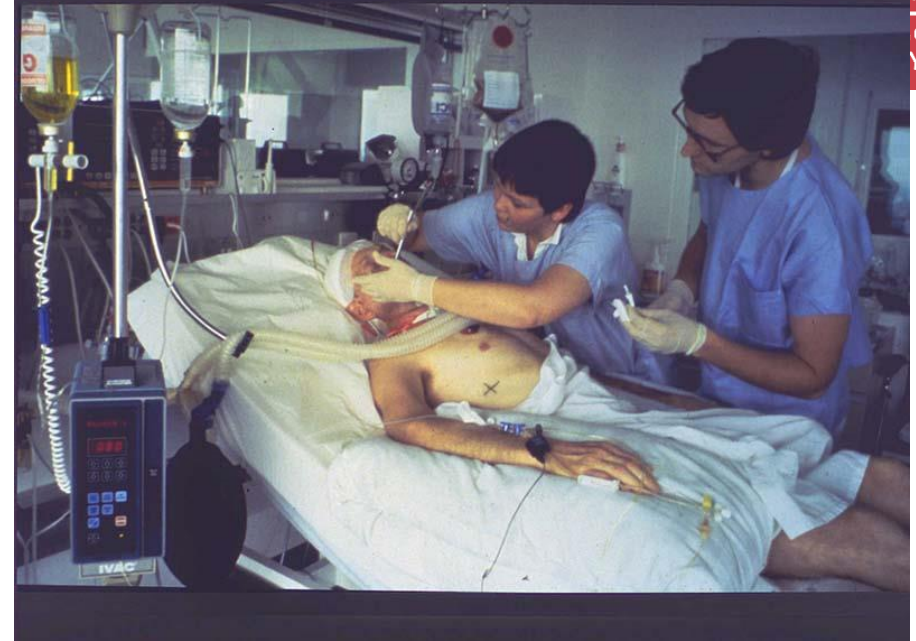
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# Modern medicine provides new opportunities for bacteria



- New entry portals
- Catheters/surgery
- Survival of the unfit
- Immuno-suppression
- Cancer R<sub>x</sub> / transplants
- Vulnerable clustered in hospital

# Why is this NICE guideline important?

surgical site infections are the cause of 20% of all of healthcare-associated infections (HCAIs) and at least 5% of patients undergoing a surgical procedure develop a surgical site infection (SSI)



**one third of all postoperative deaths attributable, at least in part, to SSIs**



# Wound Types

Impact of Wounds

Surgical site infection at 30 days post op

		<u>Pts</u>	<u>Infection</u>
Platt '90	Breast	606	7.1%
Bailey '92	Hernia	510	8.9%
Risberg '95	Vascular	580	9.1%
Taylor '97	Hernia	563	8.9%
Mellings '02	Mixed	350	14.1%

# Wound Infection Definition

## *IWII 2016*



A wound infection is the invasion of a wound by proliferating microorganisms to a level that invokes a local and/or systemic response in the host. The presence of microorganisms within the wound causes local tissue damage and impedes wound healing

# What are the challenges for the clinician?

- Diagnosis of infection
- Culturing the wound
- Managing the bioburden
- Managing the exudate
- Preventing further wound healing delay and breakdown
- Patient quality of life issues



# Wound Infection

- **Cost to heal a chronic ulcer \$40000**

Joint Commission on Accreditation of Health Care Organisations 2006

- **Cost of Single Infection**
- **HAI \$13973**
- **MRSA \$ 20891**
- **SSI \$ 15646**
- **Chronic wound \$11809**
- **Burns \$102518**

Am J Inf. Control 2002

# Risk factors for wound infection

- Systemic

- Diabetes mellitus
- Vascular disease
- Immune disorders
- Malnutrition
- Oedema
- Pharmacotherapy
- Alcoholism
- Prior Surgery

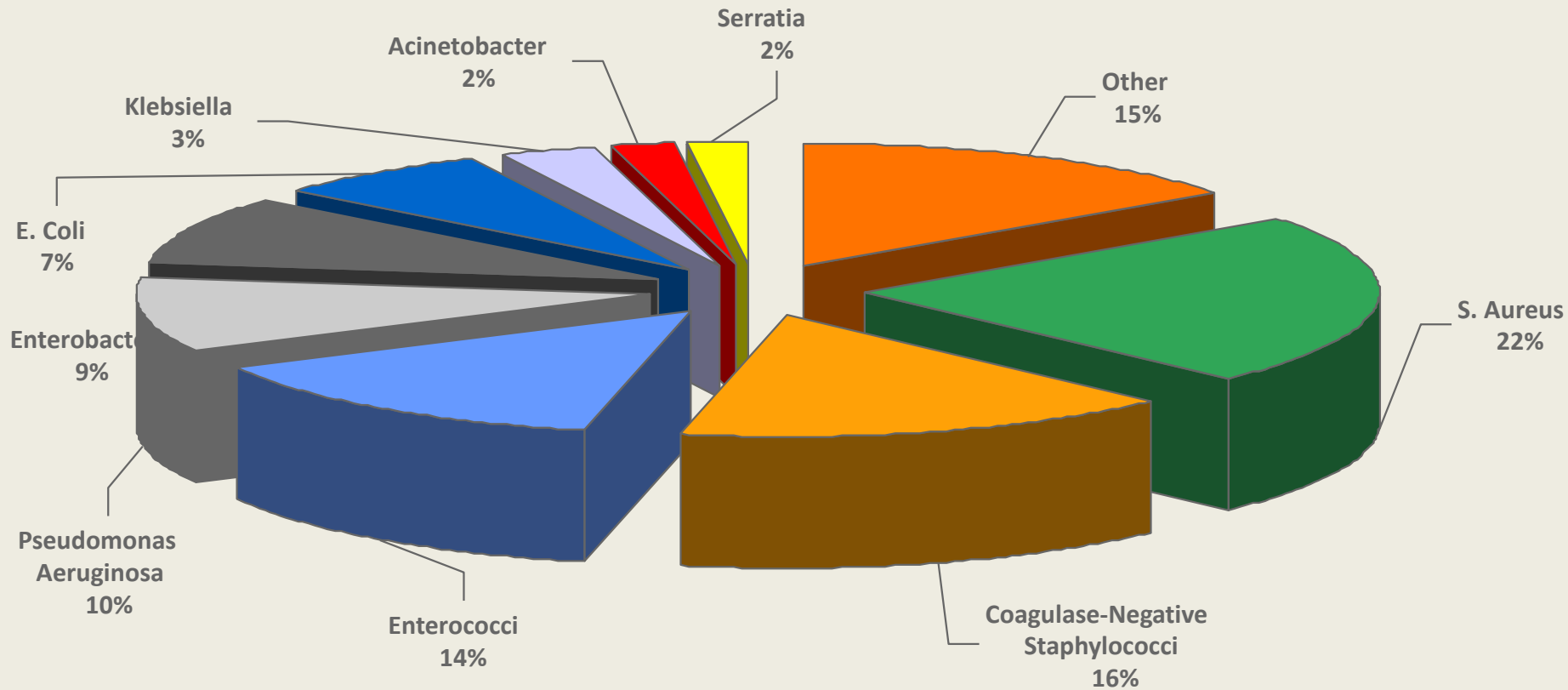
- Local

- Mechanism of injury
- Foreign body
- Reduced tissue perfusion
- Degree contamination
- Duration of wound
- Size and depth of wound
- Presence necrotic tissue



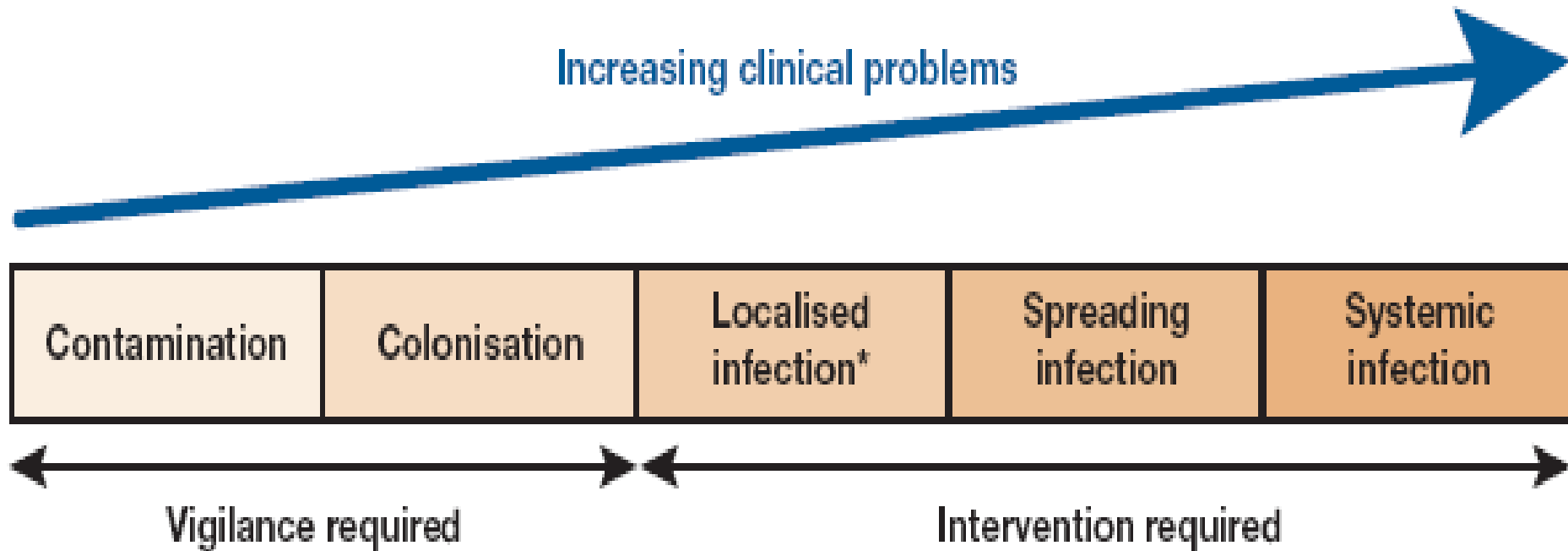
# Importance of Pathogens in the Hospital Setting

## Surgical Site Infections



*Gaynes. R. et.al CID 2005; 41: 848.*

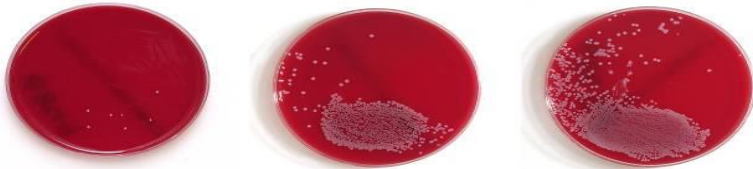
# Bacterial Burden considerations in chronic wounds



\*Localised infection may or may not be accompanied by the classical signs and symptoms of inflammation. When it is not, various terms have been used, eg critical colonisation

*World Union of Wound Healing Societies (WUWHS). Principles of best practice: Wound infection in clinical practice. An international consensus. London: MEP Ltd, 2008. Available from <http://www.woundsinternational.com/clinical-guidelines/wound-infection-in-clinical-practice-an-international-consensus>*

# Issues around diagnosis of Infection - Personal thoughts!



"Now what seems to be the problem?"



**Bacteria are nothing the terrain is everything  
- Louis Pasteur**

Infected?



Infected?





Infected ?



Infected ?





# Diagnosis of Infection

## Local Infection

- Abnormal granulation tissue
- Bleeding
- Pain \*
- Odour
- Bridging
- Delayed healing \*

*Cutting + Harding 1994*

*Gardner et al 2001*

# Diagnosis Of Soft Tissue Infection

## Sampling

- Surface culture
- Fine needle aspirates
- Curettage
- Tissue biopsy



# Optimal Technique for sampling using a swab- Can you be confident this is always done?

- Clean wound without disinfectants
  - removal of slough by cleansing
  - removal of anti-microbial substances
- Wipe with dry gauze
- Wait 1-2 minutes for interstitial fluid to collect in the base of the wound
- **Take specimen**

# Diagnosis of Infection –Gold Standard

## Tissue biopsy

95 patients' surgical wounds

If closure delayed until

$<10^5$  bacteria / gm tissue

96% success<sup>4</sup>

*Robson MC and Heggars JP (1970) J Surg Onc;2(4):379-383.*

# What is the value of microbiological diagnostics ?

The sole reporting of genus and species is only a limited information and not always the answer to the question clinician wants to know.

It is important that you know what you need to know in order to make a clinical decision!

**Most important question: Will a certain result change clinical management of a patient.**

**Treat your patients, Never treat a microbiological report**

# Lipsky, AIM 1990

- Healing 75%
- Further treatment and healing 15%
- Failed 9%
- No recurrence 84% (15 months)
- **25% grew organisms resistant to antimicrobial prescribed but wound healed**

# Swab Versus Biopsy for Chronic Wounds

- Systematic review

2332 papers found from title

49 papers found from abstract

18 partial consensus from 3 authors

3 unanimous consensus

6 papers in total agreed by 4 authors

- Biopsy result used as Gold Standard

- Levine better than Z in acute wounds
- Threshold  $3.7 \times 10^4$
- Sensitivity 0.90
- Specificity 57%
- PPV 0.77
- NPV 0.91
- Still need to optimise and standardise swabbing technique

- *Rondas et al Adv Skin & Wound Care 2013*



# Factors to consider in Wound infection

- The host, the wound, the environment
- Are wounds sterile?
- Wound swabs
- Levine technique – cleanse, rotation
- Assess – patient, blood results, clinical indicators, pain, wound bed, odour, an increase in exudate

***Do not undertake microbiological analysis of wound specimens in the absence of an appropriate indication (IWII 2017)***

# Wound Swab Misuse in Practice

- Wound swab, patient co-morbidities, wound description, white cell count, CRP, antibiotic prescriptions
- 134 patients, **diabetes, post-op wounds**
- Swab results not acknowledged in case notes in 76% of cases
- Wound swabs – not indicated, poor documentation, untimely acknowledgment of results

*Fenech et al 2014*

# Routine Culture of Leg ulcers

## Cardiff Study

- 178 patients sampled for 12 weeks
- 153 species identified
- Staph aureus 64.3%
- C. Striatum 60.6%
- Pseudomonas 32.6%
- No correlation between species or anaerobes and healing

## Clinical Appearances

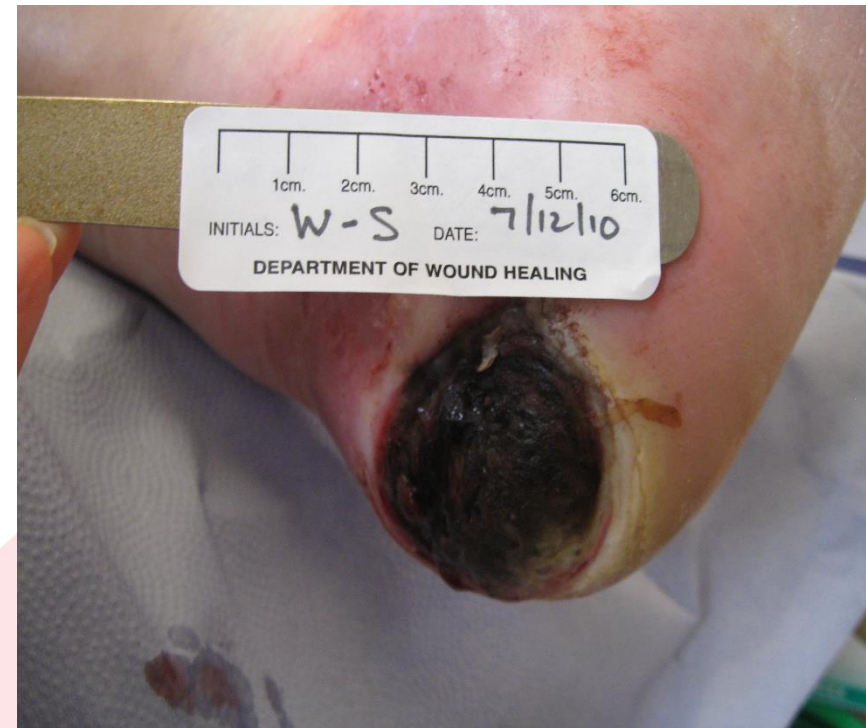


# Anaerobes and Infection in ulcers

## Challenges that exist

- 220 isolates from 44 infected ulcers
- 110 isolates from 30 non infected ulcers
- Greater number of anaerobe species in infected ulcers 2.5 vs 1.3
- Anaerobes 49% of infected and 36% of non infected
- Staph aureus more common in non infected rather than infected ulcers

## Clinical picture



# Clinical Signs of Infection in Chronic Wounds

## Difficult to Diagnose

- 36 Chronic wounds
- Signs & symptoms of Infection
- Interobserver variability 0.53-1.0
- Cultured only 31% infected
- Positive prediction by clinical signs
- Sensitivity 0.62 clinical
- Sensitivity 0.38 classical

## Infected?



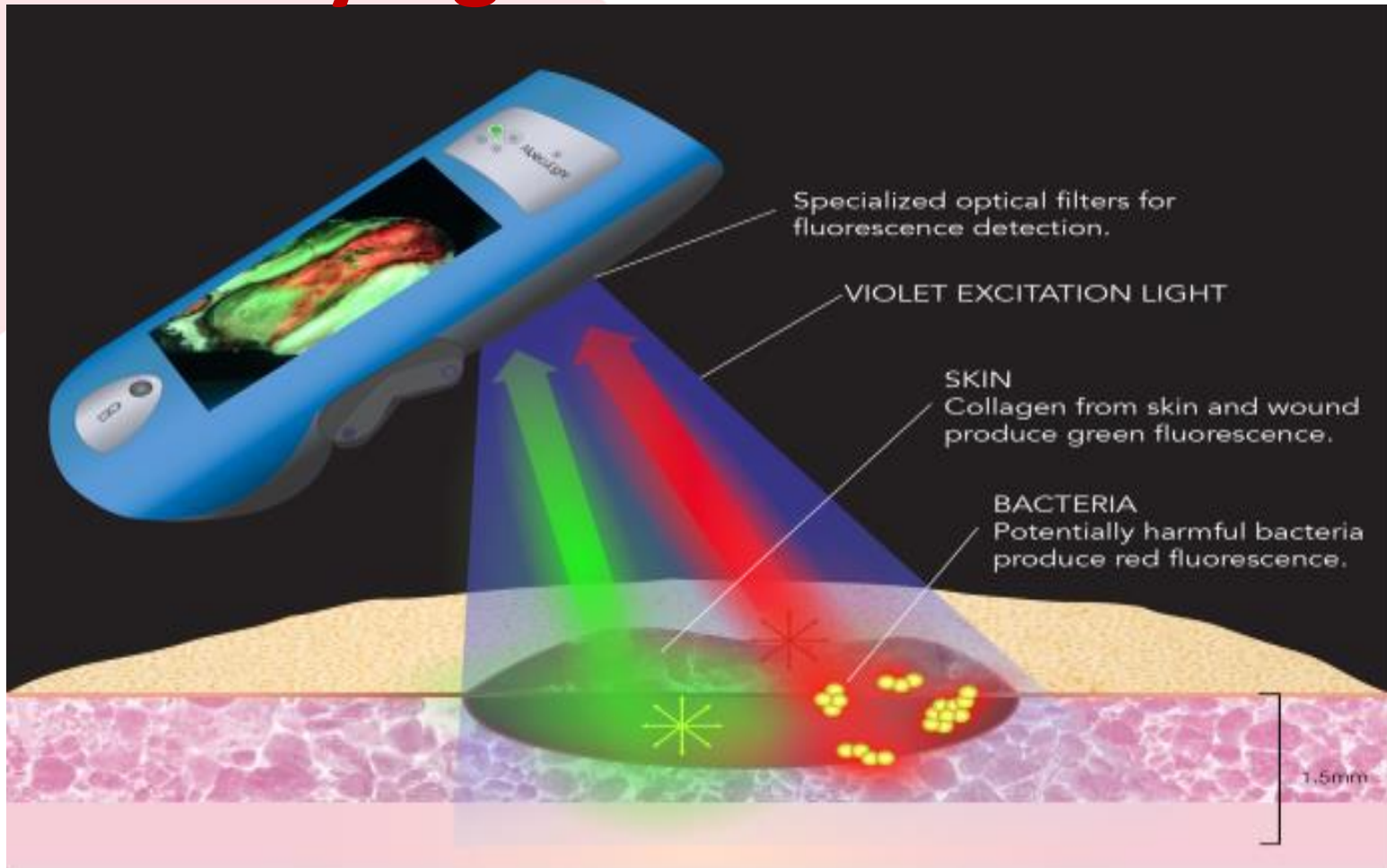


# Pain as a symptom of infection in wounds

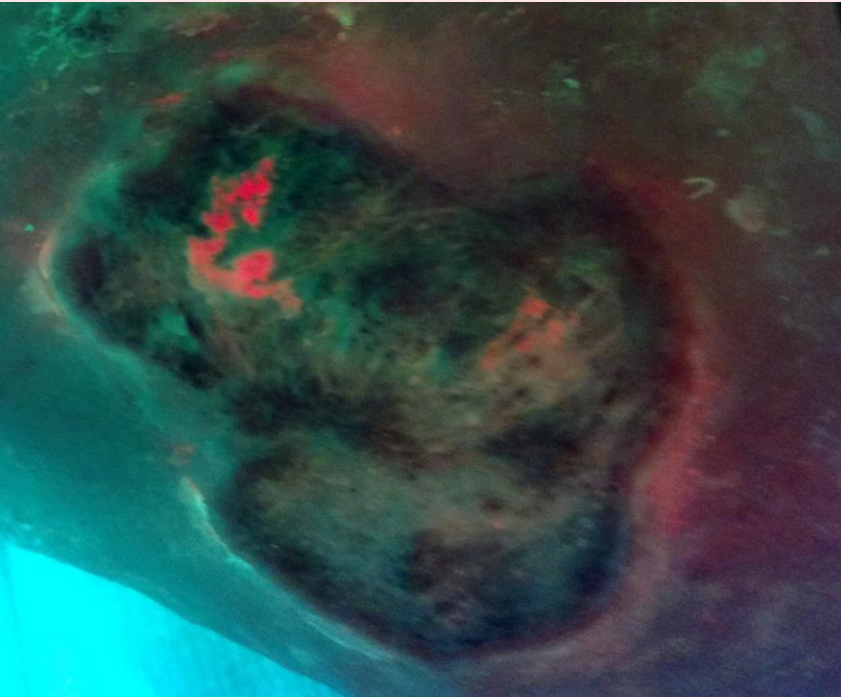
- Systematic Review to Nov 2011
- 341 studies screened
- 15 basis of review
- Includes 985 patients and 1056 wounds
- Prevalence infection 53%
- In Infected wounds
- Increase in pain Likelihood ratio 11-20
- Absence of pain 0.64 -0.88
- Examination and lab tests limited value

*Reddy et al JAMA 2012*

# How accurate are clinicians at identifying location of bacteria?



# HOW CONFIDENT AND HOW ACCURATE?



**18 Clinicians 90 images**

**62% Wound Experts**

**79% of images not maximum site of bacteria**

**Confidence Score of 6.9 (1-10)**

**Clinical Usefulness Score of 8.2 (1-10)**

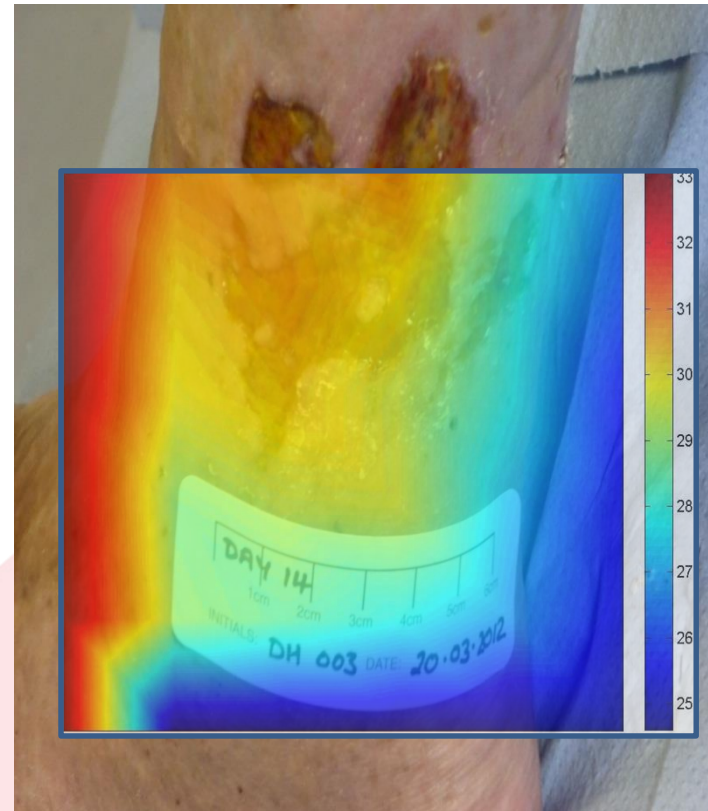
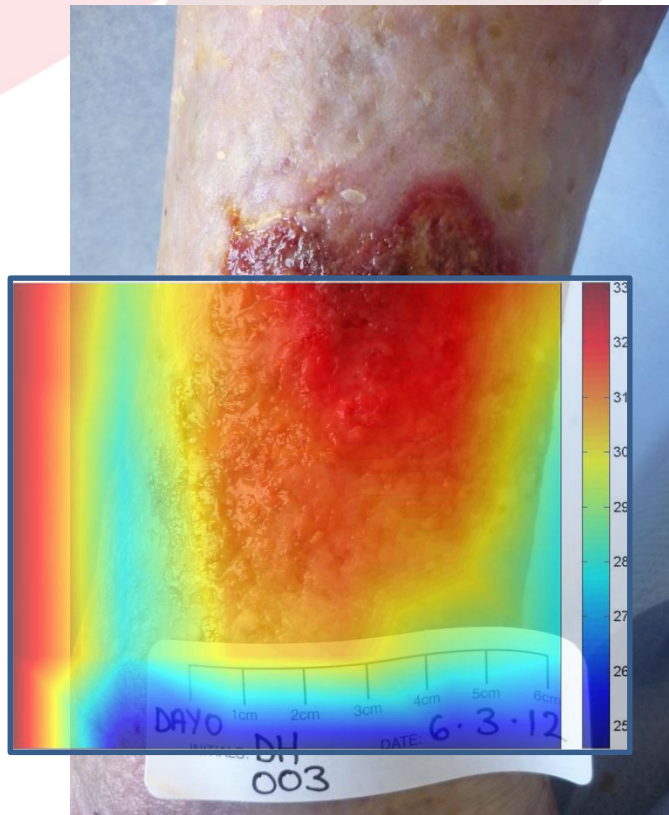
***Naik FIS 2017***

# Clinical diagnosis of infection

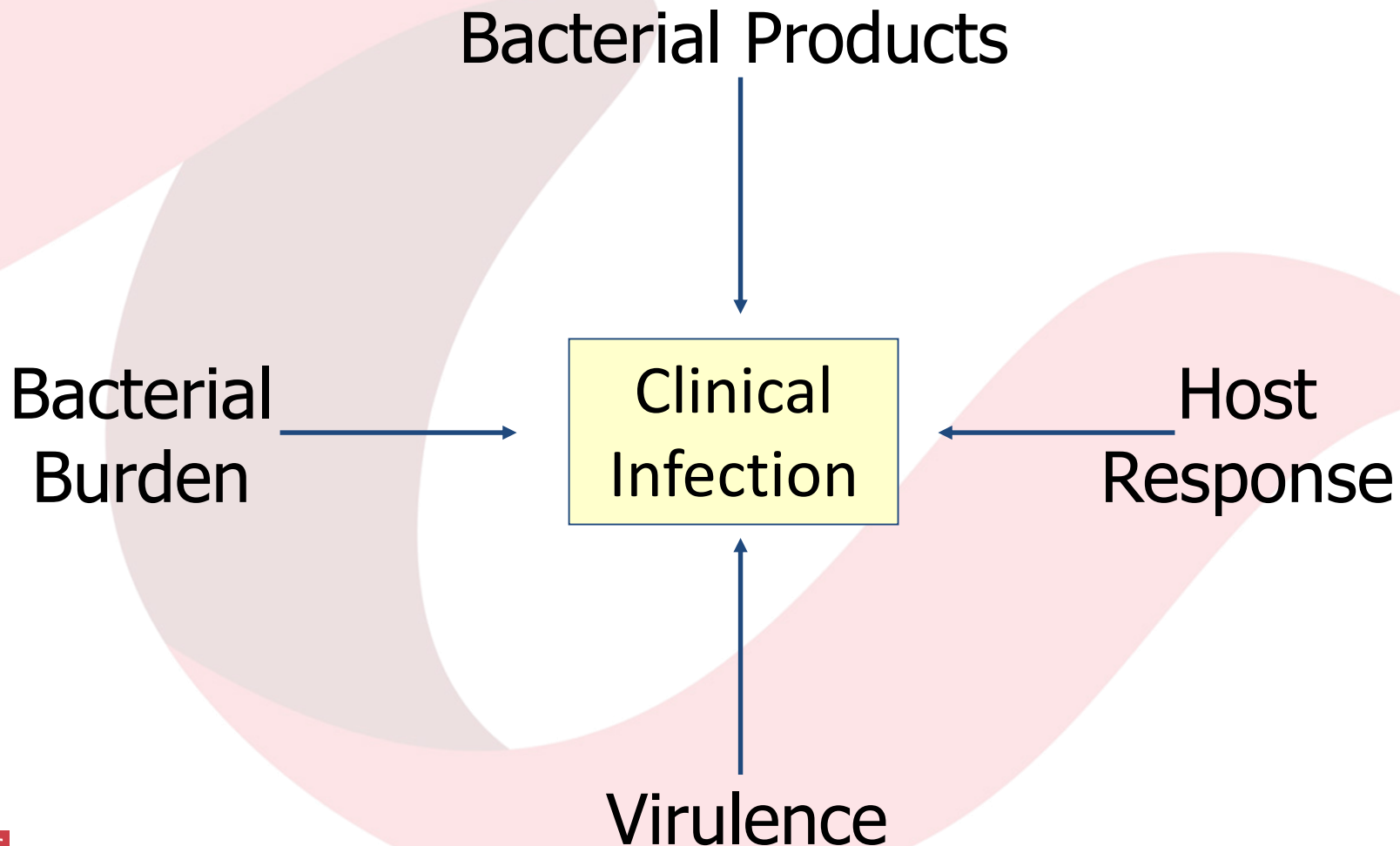




# Temperature mapping in chronic wounds



# Colonisation or Infection

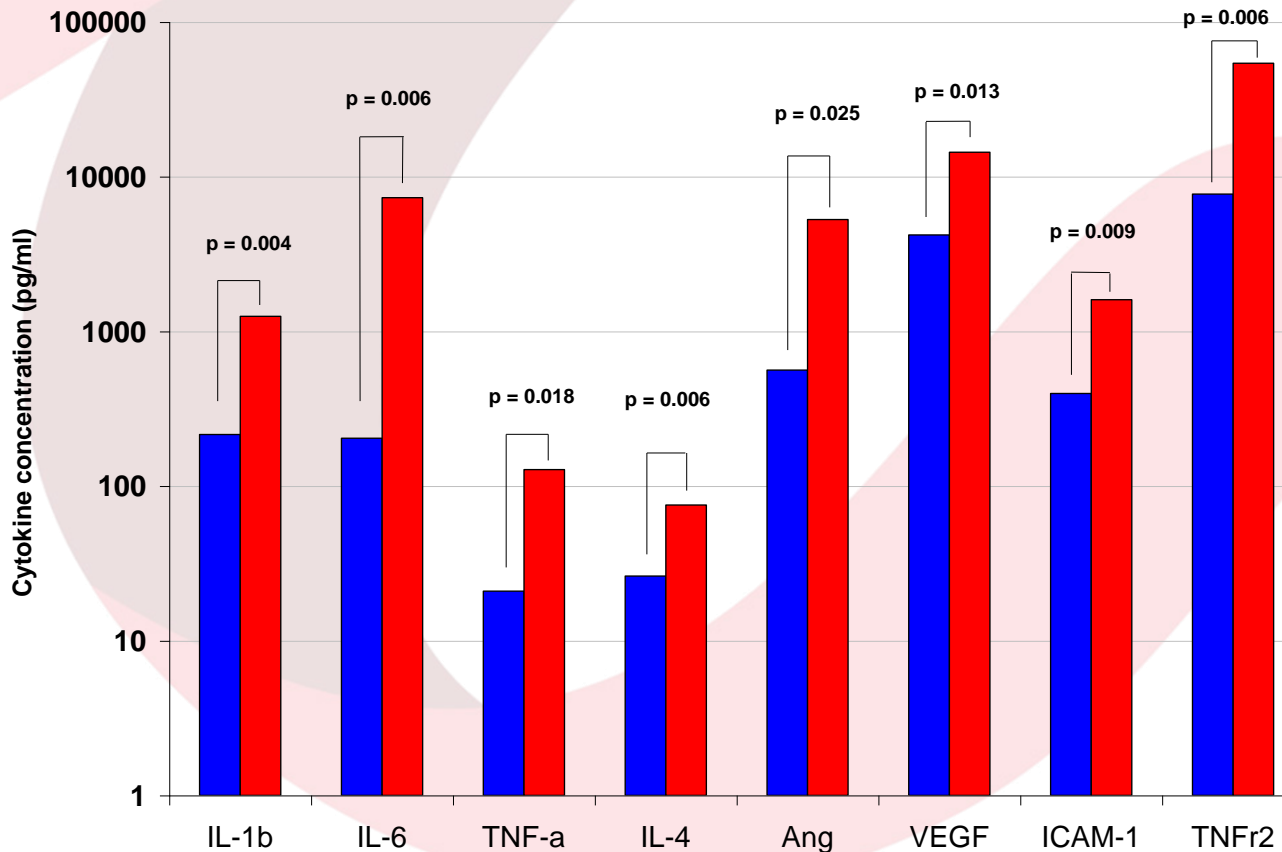


*Williams CID 2004*



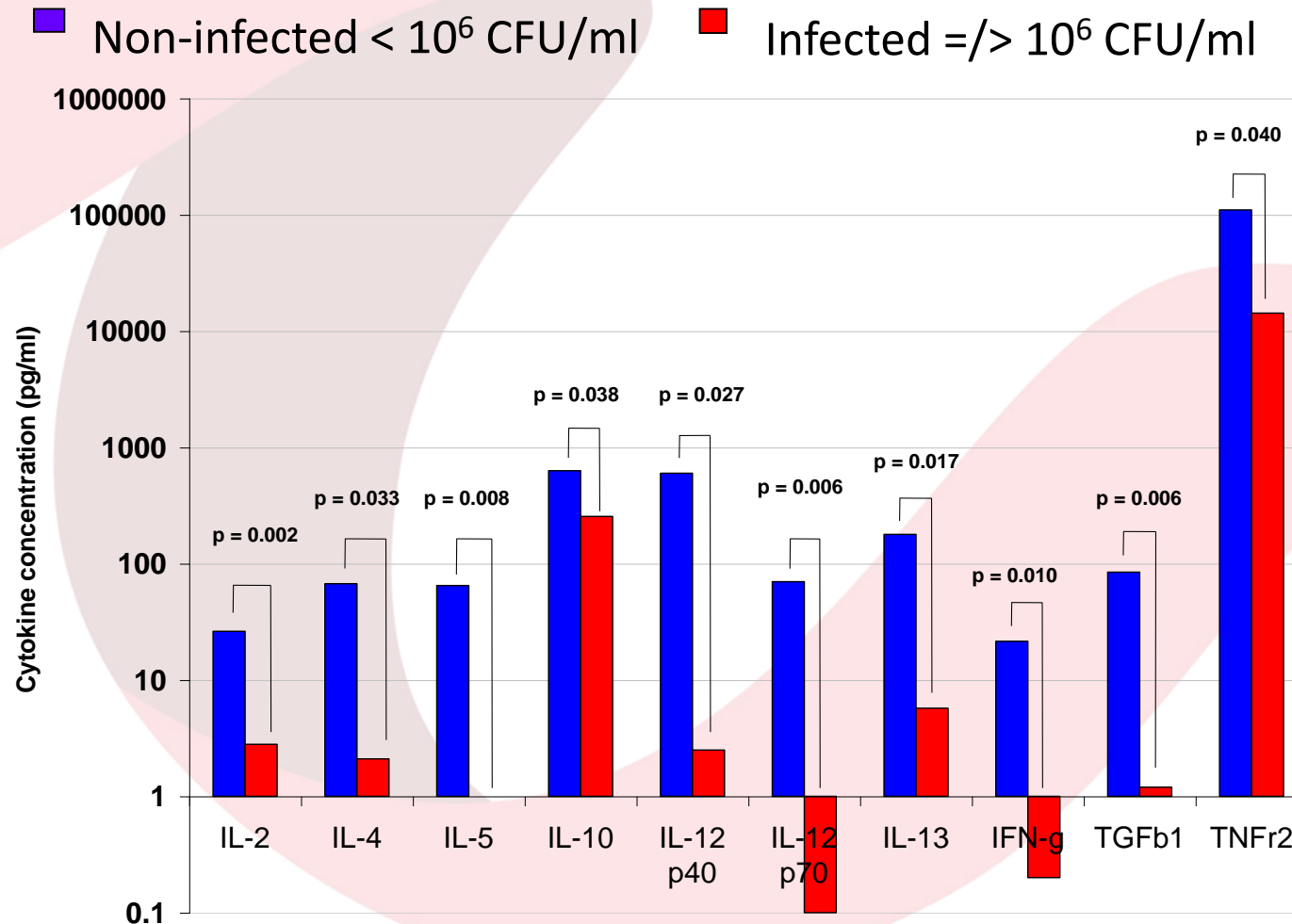
# Cytokine Levels in VLU & Microbial Diagnosis

■ Non-infected < 10<sup>6</sup> CFU/ml ■ Infected =/> 10<sup>6</sup> CFU/ml



Clark et al 2014 WRR

# Cytokine Levels in DFU & Microbial Diagnosis



Clark et al 2014 WRR

# Maintaining a high level of clinical suspicion

- Wound infection is a common complication
  - At least 5% of patients develop infection after a surgical procedure
  - Around 50% of chronic wounds may be infected
  - Around 60% of chronic wounds contain biofilm
- **Diagnosis is primarily based on clinical assessment of signs and symptoms**

# Chronic Wounds: Advanced Wound Dressings and Anti-microbial dressings

- Dressings provide optimal environment for healing
- They work by physical or chemical means typically by controlling moisture levels
- Few RCT's
- Many low quality
- Effects are uncertain
- Use cheapest dressing
- Consider frequency of dressing change
- Silver should not be used unless clinical signs or symptoms of infection are present

*NICE guidance 2016*

# Treatment of Infection

Modern day antiseptics

- Iodine
- Cadexomer iodine
- Silver nitrate (?)
- Silver Sulfadiazine
- Nanocrystalline silver
- Other silvers
- PHMB

**Ask 2 simple questions of any topical antibacterial agent**

**1. Show me it reduces bacteria in a wound**

**2. Show me it can help reduce infection rates**



# Evidence for therapeutic interventions in infection

- Topical antiseptic agents <sup>7-10</sup>
- Antimicrobial therapy <sup>11-12</sup>
- Systemic antibiotics <sup>13</sup>
- Surgical debridement <sup>14-15</sup>
- Topical negative pressure <sup>16</sup>
- Granulocyte-colony stimulating factor <sup>17</sup>



- **7. Martínez-De Jesús FR, Ramos-De la Medina A, Remes-Troche JM, et al. Efficacy and safety of *neutral pH superoxidised solution* in severe diabetic foot infections. *Int Wound J* 2007; 4: 353–362.**
- **8. Chen W, Xu K, Zhang H, Shang Y, Hao P. A comparative study on effect of bacterial load in diabetic foot ulcers dealing with *iodophor and rivanol* respectively. *Zhongguo Xiu Fu Chong Jian Wai Ke Za Zhi* 2008; 22: 567–570.**
- **9. Piaggese A, Goretti C, Mazzurco S, et al. A randomized controlled trial to examine the efficacy and safety of a new *super-oxidized solution* for the management of wide postsurgical lesions of the diabetic foot. *Int J Low Extrem Wounds* 2010; 9: 10–15.**
- **10. Landsman A, Blume PA, Jordan DA Jr, Vayser D, Gutierrez A. An open-label, three-arm pilot study of the safety and efficacy of *topical Microcyn* Rx wound care versus oral levofloxacin versus combined therapy for mild diabetic foot infections. *J Am Podiatr Med Assoc* 2011; 101: 484–496.**
- **11. Lipsky BA, Peters EJ, Senneville E, et al. Expert opinion on the management of infections in the diabetic foot. *Diabetes Metab Res Rev* 2012; 28(Suppl 1): 163–178.**
- **12. NICE. Chronic wounds: *advanced wound dressings and antimicrobial dressings*. Available at: <https://www.nice.org.uk/advice/esmpb2/chapter/Evidence-review>, NICE Advice ESMPB2, March 2016.**

- 13. Lipsky B.A, Aragon-Sanchez J, Diggle M, Embil J, Kono S, Lavery L, Senneville E, Urbancic-Rovan V, Van Asten s, Peters E. J. G. IWGDF 2015 Guidance on the diagnosis and management of foot infections in persons with diabetes. Available at: <http://iwgdf.org/guidelines/guidance-on-infection/>
- 14. Tan JS, Friedman NM, Hazelton-Miller C, Flanagan JP, File TMJ. Can **aggressive treatment of diabetic foot infections** reduce the need for above-ankle amputation? Clin Infect Dis 1996; 23: 286–291.
- 15. Faglia E, Clerici G, Caminiti M, Quarantiello A, Gino M, Morabito A. The role of **early surgical debridement and revascularization in patients with diabetes** and deep foot space abscess: retrospective review of 106 patients with diabetes. J Foot Ankle Surg 2006; 45: 220–226.
- 16. Dalla Paola L, Carone A, Ricci S, Russo A, Ceccacci T, Ninkovic S. Use of **vacuum assisted closure therapy** in the treatment of diabetic foot wounds. J Diabetic Foot Complications 2010; 2: 33–44.
- 17. Cruciani M, Lipsky BA, Mengoli C, de Lalla F. **Granulocyte-colony stimulating factors** as adjunctive therapy for diabetic foot infections. Cochrane Database of Sys Rev3 2013; 8: 1–30. CD006810. DOI:10.1002/14651858.CD006810.pub3.

# Getting the basics right: TIME

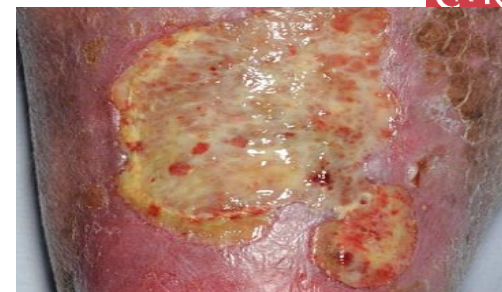
(Schultz et al. 2003)

## Wound bed preparation:

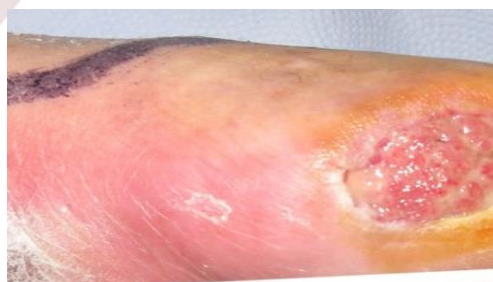
*"The global management of the wound to accelerate endogenous healing or to facilitate the effectiveness of other therapeutic measures."*

*Falanga, 2002*

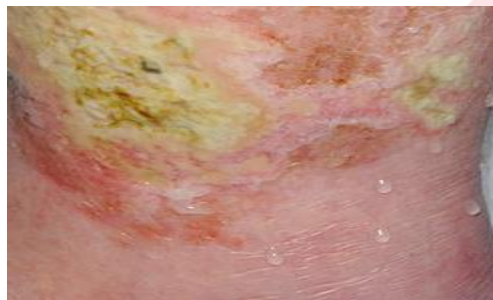
Tissue



Infection /  
Inflammation



Moisture  
imbalance



Edge



# Tissue





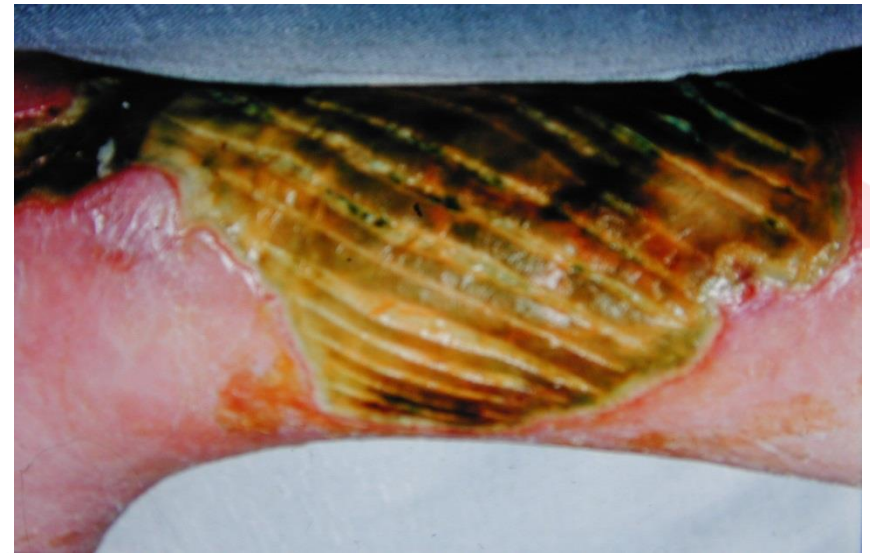
# Why debride?

Devitalised tissue:

- May mask/mimic signs of wound infection
- Is a source of nutrients for bacteria
- Is a physical barrier to healing and topical therapies
- Prolongs the inflammatory process
- Hinders wound assessment

# EWMA definition of debridement

The act of removing necrotic material, eschar, devitalised tissue, serocrusts, infected tissue, hyperkeratosis, slough, pus, hematomas, foreign bodies, debris, bone fragments or any other type of bioburden from a wound with the objective to promote wound healing





# Mechanical Debridement - New developments

## Pad

- All wound types, surface
- 18M angled fibre tips loosen debris and exudate
- Safe fixation of fibres
- Reduction of linting
- High stability
- Reverse: polyacrylate



## Lolly

- Cavity wounds, surgical
- Hard-to-reach areas
- 8M angled fibres
- Securely fixated head
- X-ray detectable thread
- Easy-grip handle
- Visual orientation



# Which of these wounds are infected or inflamed?



# Moisture Balance



Edge





# The reality of Diabetic foot disease





# Guidelines for diagnosis and management of diabetic foot infections

Based on a Systematic Review of the Effectiveness of Interventions in the Management of Infection in the Diabetic Foot' and 'Expert Opinion on the Management of Infections in the Diabetic Foot'.

## Diagnosis

- Every diabetic patient with a foot wound should be assessed for the presence of infection.
- **The diagnosis of diabetic foot infection is based on clinical findings of inflammation, rather than solely the results of culture.**
- The severity of infection should be assessed after debridement of callus and necrotic tissue on the basis of its extent and depth and the presence of any systemic inflammatory findings.
- Hospitalisation is needed for all patients with a severe infection, many patients with a moderate infection but few with mild.

Lipsky et al Specific guidelines for the treatment of diabetic foot infections (2012) Diabetes Metab Res Rev; 28(Suppl 1): 234–235.

# Recommendation: classification & diagnosis

**Table 1.** *The classification systems for defining the presence and severity of an infection of the foot in a person with diabetes developed by the Infectious Diseases Society of America (IDSA) and the infection part of the PEDIS classification of the International Working Group on the Diabetic Foot (IWGDF) (29,30).*

Clinical classification of infection, with definitions	IWGDF / IDSA classification
<b>Uninfected:</b> No systemic or local symptoms or signs of infection	1 (Uninfected)
<b>Infected:</b> <ul style="list-style-type: none"> <li>- At least 2 of the following items are present: <ul style="list-style-type: none"> <li>• Local swelling or induration</li> <li>• Erythema &gt; 0.5 cm* around the wound</li> <li>• Local tenderness or pain</li> <li>• Local warmth</li> <li>• Purulent discharge</li> </ul> </li> <li>- Other causes of an inflammatory response of the skin should be excluded (e.g., trauma, gout, acute Charcot neuro-osteoarthropathy, fracture, thrombosis, venous stasis)</li> <li>- Infection involving only the skin or subcutaneous tissue (without involvement of deeper tissues and without systemic manifestations as described below).</li> <li>- Any erythema present extends &lt; 2 cm* around the wound</li> <li>- No systemic signs or symptoms of infection (see below)</li> </ul>	2 (Mild infection)
<ul style="list-style-type: none"> <li>- Infection involving structures deeper than skin and subcutaneous tissues (e.g., bone, joint, tendon, muscle) or erythema extending &gt;2 cm* from the wound margin.</li> <li>- No systemic signs or symptoms of infection (see below)</li> </ul>	3 (Moderate infection)
<ul style="list-style-type: none"> <li>- Any foot infection with the systemic inflammatory response syndrome (SIRS), as manifested by ≥2 of the following: <ul style="list-style-type: none"> <li>• Temperature &gt;38° or &lt;36° Celsius</li> <li>• Heart rate &gt;90 beats/minute</li> <li>• Respiratory rate &gt;20 breaths/minute or PaCO<sub>2</sub> &lt; 4.3 kPa (32 mmHg)</li> <li>• White blood cell count &gt;12,000 or &lt;4,000/mm<sup>3</sup>, or &gt;10% immature (band) forms</li> </ul> </li> </ul>	4 (Severe infection)

**Note:** \*In any direction, from the rim of the wound; The presence of clinically significant foot ischemia makes both diagnosis and treatment of infection considerably more difficult.

*IWGDF Guidance on the diagnosis and management of foot infections in persons with diabetes*

# IWGDF Infection Review 2015

Papers published up to 30 June 2014

37 RCTs

3 Cohort studies

40 in total

Of these:

15 RCTs – antibiotics for skin and soft tissue

10 RCTs – antibiotics for osteomyelitis

Others:

Surgical procedure

Topical antiseptics

NPWT

HBO

## Conclusions

**No clear guidance possible, rely on expert opinion**

*Diab Met Rev 2015*

Department of Health  
Advisory Committee on Antimicrobial Resistance  
and Healthcare Associated Infection (ARHAI)

# ANTIMICROBIAL STEWARDSHIP: "START SMART - THEN FOCUS"

Guidance for antimicrobial stewardship  
in hospitals (England)

*Davies J, Davies D. Origins and Evolution of Antibiotic Resistance. Microbiology and Molecular Biology Review. September 2010. Vol 74 (3): 417-433*

*Zimlichman E, Daniel Henderson D, Tamir O, Franz C, Song P, Yamin C.K, Keohane C, Denham C.R, MD<sup>6</sup>;*

*Bates D.W. Health Care–Associated Infections: A Meta-analysis of Costs and Financial Impact on the US Health Care System. JAMA Intern Med. 2013;173(22):2039-2046.*

# Antibiotic Stewardship in Wounds

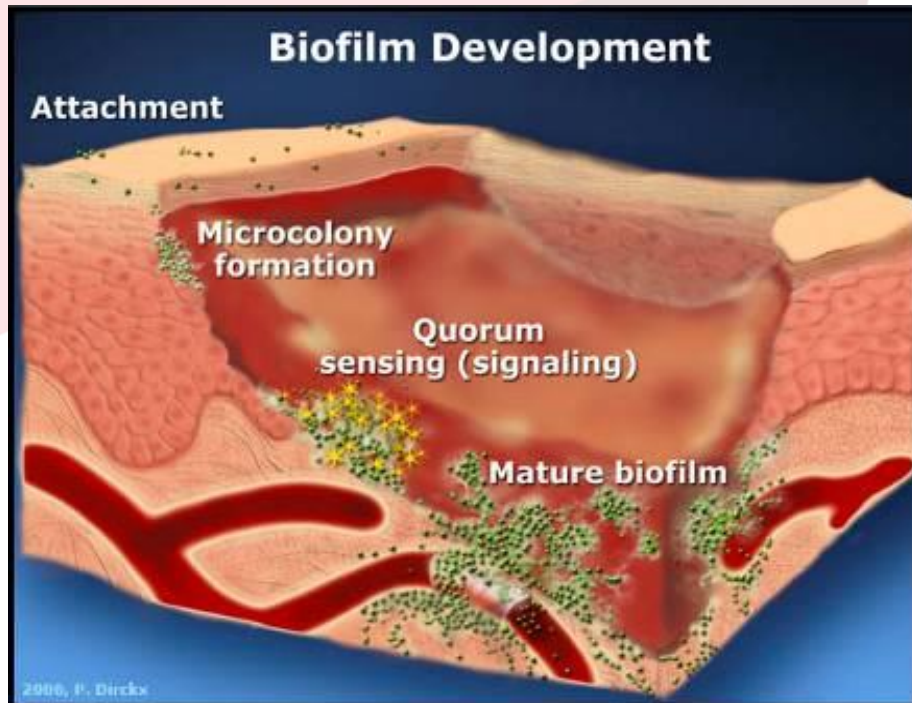
## Factors contributing to Antimicrobial Misuse

- Diagnostic Uncertainty
- Clinical Ignorance
- Clinician Fear
- Patient Demand

*Lipsky et al JAC 2016*



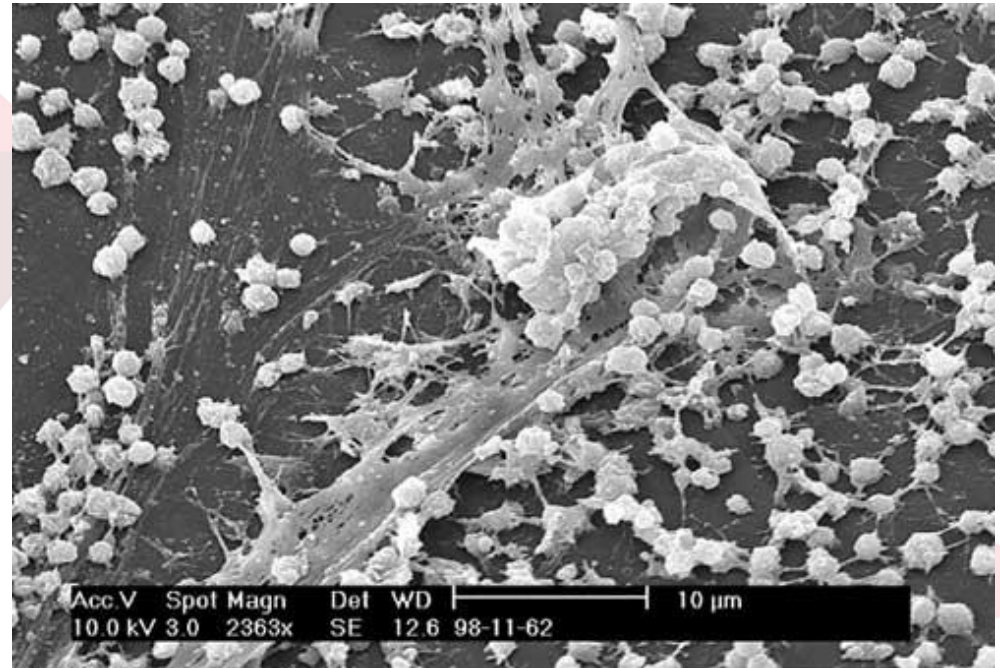
# Biofilms



- The presence of a biofilm perpetuates the chronic inflammation by recruiting neutrophils and macrophages, changing macrophages' phenotype to be pro-inflammatory and have reduced microbicidal activity, and increasing inflammatory cytokine levels (e.g. Interleukins, TNF $\alpha$ )

*Zhao G et al. Biofilms and inflammation in chronic wounds. Adv Wound Care 2013;2(7):389-399.*

# Definition of Biofilm



A **structured community** of bacterial cells enclosed in a self-produced polymeric **matrix** and **adherent** to an inert or living surface<sup>6</sup>.

# Definition of a Biofilm

## *IWII 2016*



*A structured community of microbes\* with genetic diversity and variable gene expression (phenotype) that creates behaviours and defences used to produce unique infections (chronic infection). Biofilms are characterised by significant tolerance to antibiotics and biocides while remaining protected from host immunity.*

*\* Microbes to replace the term bacteria as organisms other than bacteria (e.g. fungi) are common causatives of wound infection.*

# Biofilms

- A biofilm is bacteria embedded in a thick slimy barrier of sugars and proteins  
[http://www.woundsinternational.com/pdf/content\\_8851.pdf](http://www.woundsinternational.com/pdf/content_8851.pdf)
- Attach to living or non living surface
- Single or polymicrobial
- Microscopic

# Biofilm Staging

- Stage 1 – reversible surface attachment (planktonic bacteria)
- Stage 2 – permanent surface attachment, (quorum sensing)
- Stage 3 – slimy protective matrix/biofilm, (extra cellular polymeric substance, EPS)
- Maturation and dispersal



# Biofilm Issues

- Mature biofilms shed planktonic bacteria
- Mechanical disruption
- Reform within 24 hours
- Sub population hibernate
- Chronic inflammatory response

# Potential role of Biofilm in persistent inflammation

## Biofilms:

- Surrounded by neutrophils - do not penetrate / destroy them
- Recruit neutrophils and macrophages to wound environment
- Change macrophage phenotype to be pro-inflammatory and have reduced microbicidal activity
- Increase inflammatory cytokine levels (e.g. Interleukins, TNF $\alpha$ )
- May induce keratinocytes to produce more MMPs (in vitro evidence)

*Zhao et al. 2013*

# Biofilms protect Bacteria

- Blocking – against antimicrobial agents and inflammatory cells
- Mutual protection – antibiotic-resistant bacteria secrete enzymes to protect other bacteria, gene transfer of resistance
- Reduced growth rate – hibernation (quiescence)
- Almost all antibiotics kill metabolically active bacteria

***Clinicians should assume that all non-healing, chronic wounds that have failed to respond to standard care have biofilm***

***(WUWHS 2016)***

# Polymicrobial infections in Chronic Diabetic wounds

- Staph Aureus most common
- Usually polymicrobial
- Staph epidermidis may become pathogenic when exposed to circulation through wound bed
- Biofilms common
- Quorum sensing that regulate genes and proteases
- Biofilms exhibit enhanced tolerance to antimicrobial agents
- Traditional antibiotics ineffective against biofilms by
  - Multi layered defense
  - Adaptive stress responses
  - Metabolic inactivation due to limited nutrients
  - Different charged surface on biofilms may influence antibiotic penetration

*Buch et al Clin Micro Reviews 2019*

# Which of these wounds has a biofilm present?





# How can I treat biofilms?

Bacteria in biofilms are difficult to kill with topical or systemic antibiotics, antibacterials, or antiseptics.

**Biofilm-based wound management involves:**

- **LOCATE and REMOVE BIOFILMS** by effective debridement techniques  
**and then**
- **Use “Biofilm busting” products**
- **PREVENT THE REFORMATION OF BIOFILMS**

# Biofilms as a cause of infection

## Facts

- Cause of 65-80% infections
- Bacteria 10-1000 X more resistant to conventional antibiotics
- No specific anti biofilm agents exist
- Small peptides may help

*J Bacteriology 2016*

## Clinical appearance



# Wound infection Continuum

## IWII 2016



WELSH WOUND  
INNOVATION  
ARLOESDD  
GLWYFAU CYMRU

BIOFILM

Increasing microbial virulence and/or numbers

Contamination

Colonisation

Local infection

Spreading infection

Systemic infection

Vigilance required

Intervention required

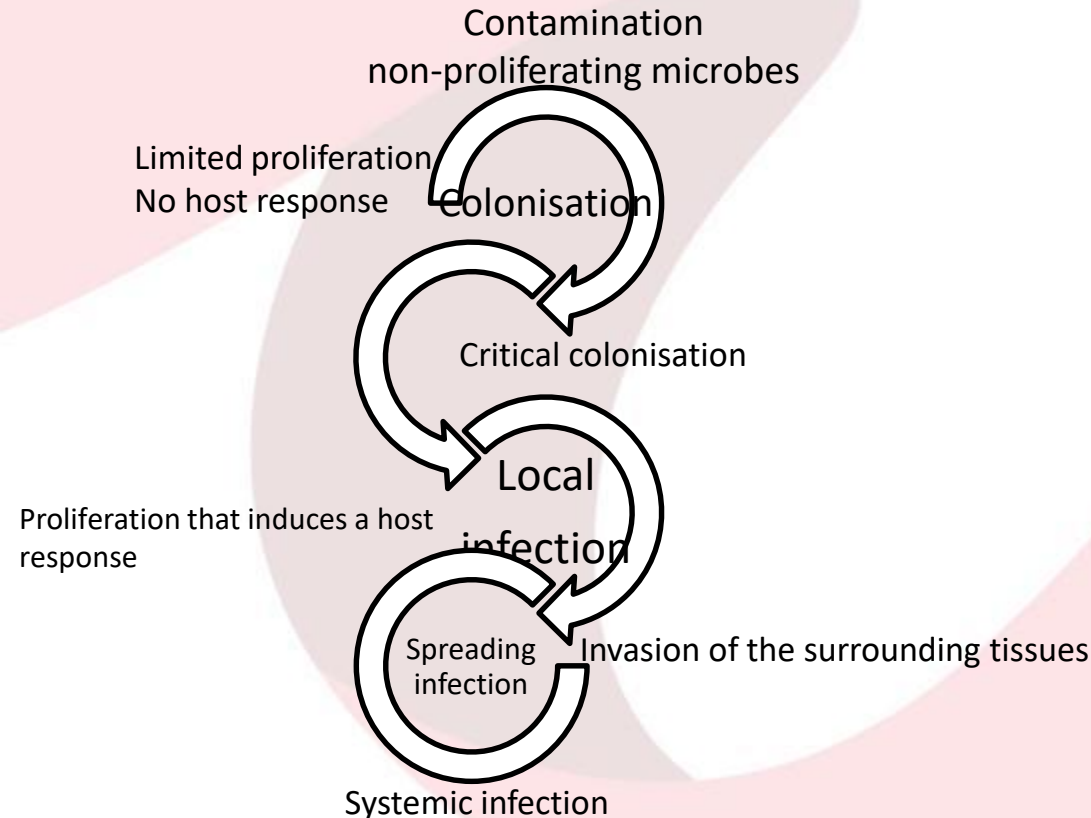
No antimicrobials indicated

Topical antimicrobial

Systemic and topical antimicrobials



# Wound Healing Continuum



## IWII 2016

- Critical colonisation first proposed in 1998
- Covert signs of infection apparent before the classic (overt) signs
- **Latest suggestion is to remove the term critical colonisation**

# Local infection

## Covert signs

- Overgranulation
- Friable/bleeding granulation
- Epithelial bridging
- Spontaneous bleeding
- Wound breakdown
- Delayed wound healing
- New/increasing pain
- New/increasing odour

## Overt signs

- Erythema
- Local warmth
- Swelling
- Purulent discharge
- Delayed healing
- New/increasing pain
- New/increasing odour
- Increase in wound exudate



# Infection



WELSH WOUND  
INNOVATION  
ARLOESIDD  
GLWYFAU CYMRU

## Spreading infection

- Extending erythema
- Lymphangitis
- Crepitus
- Wound breakdown
- Malaise/lethargy/loss of appetite
- Swelling of the lymph glands
- Increased white cell count/
- C-reactive protein (CRP)
- Pyrexia

## Systemic infection

- Sever sepsis
- Septic shock
- Organ failure
- Death

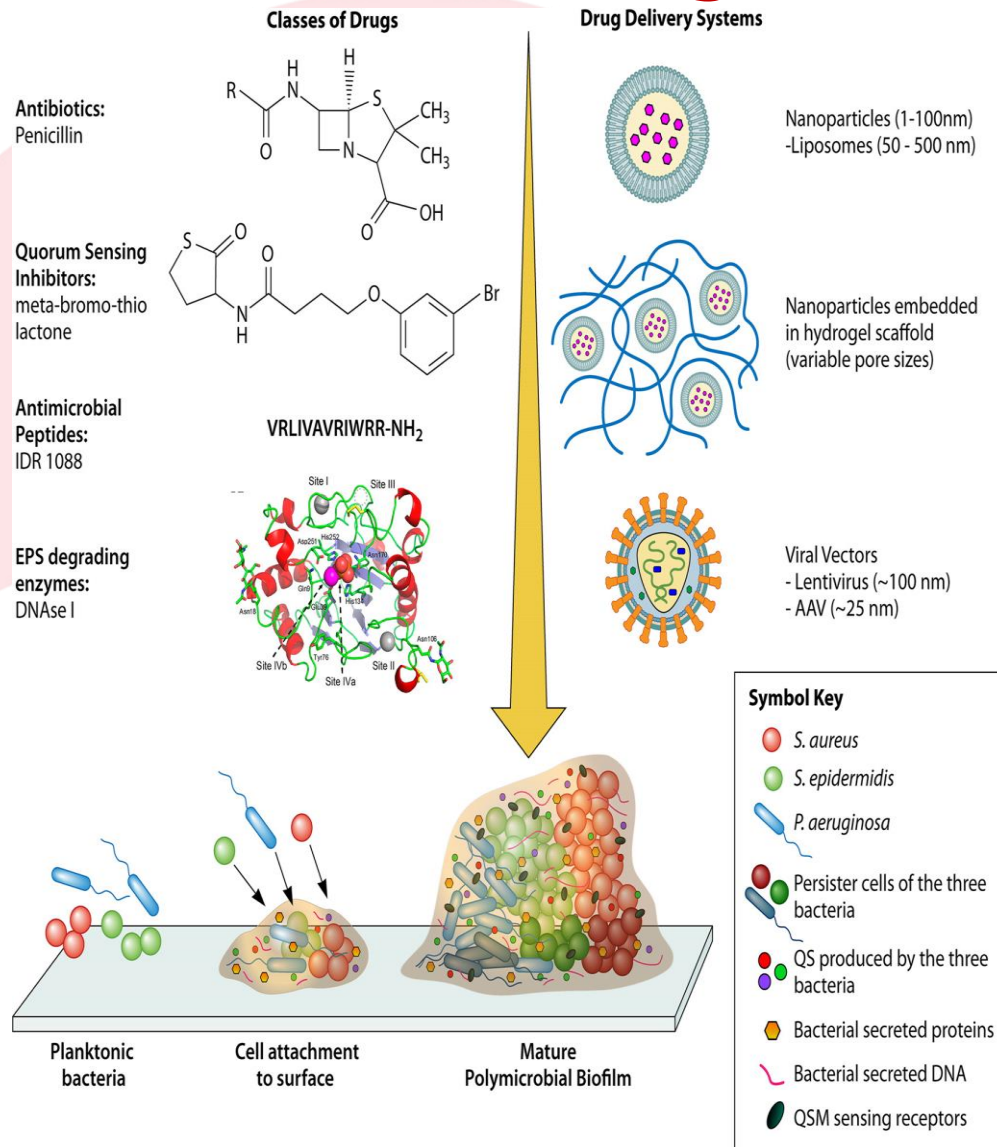
# New Test to Identify Biofilms ?



- Micro organisms release VOC's
- Identify VOC profiles of species in vitro and ex vivo
- Results influenced by model
- Some VOC's specific for planktonic or biofilm formers
- Potential for developing non invasive test in practice

*Ashrafi et al Sci Reports 2018*

# Treatment Strategies for Biofilms in Wounds



- Biofilm formation and treatment options for chronic wounds. Planktonic bacteria secrete extracellular proteins and DNA and form a glycocalyx containing polysaccharide film around them, which marks the beginning of the formation of a biofilm.
- As the number of bacterial cells in the polysaccharide matrix increases due to cell division and from the environment, the matrix thickens and forms a mature biofilm.
- Each bacterial species proliferates in its own "territory" until nutrient and gas supplies are not limiting and secretes quorum-sensing molecules.
- Several classes of drug molecules exist for treating bacterial infections, but their efficacy is limited since they either cannot penetrate the matrix or are degraded by matrix components.
- Drug delivery systems have evolved to attenuate the problem.

*Buch et al Clin Micro Reviews 2019*

# Surfactants and Biofilms

- Surfactants are surface active agents known to reduce surface tension between 2 liquids or a liquid and solid surface
- Chemically Produced Anionic/Non ionic/Cat ionic
- Biosurfactants
- Glycolipids/Lipopeptides/Fatty Acids/Phospholipids/
- Surfactant cleaners have increased healing rates
- Non Ionic surfactant poloxamer 188 can cause cell membrane repair and has antibiofilm properties

*Percival et al IWJ 2018*

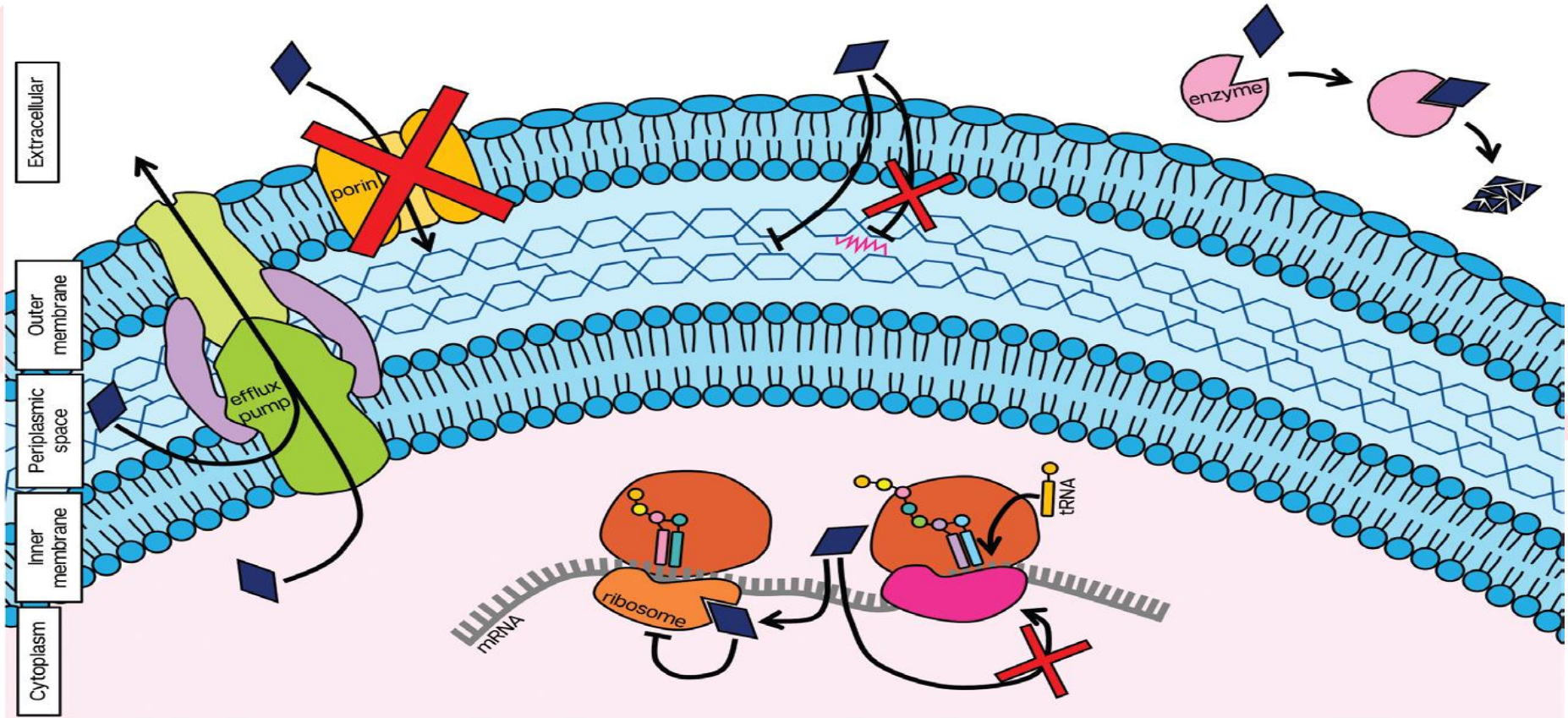


# Alternatives to Treat Infected Wounds

A) Reduction of drug accumulation

B) Modification of antibiotic targets

C) Inactivation of antibiotics

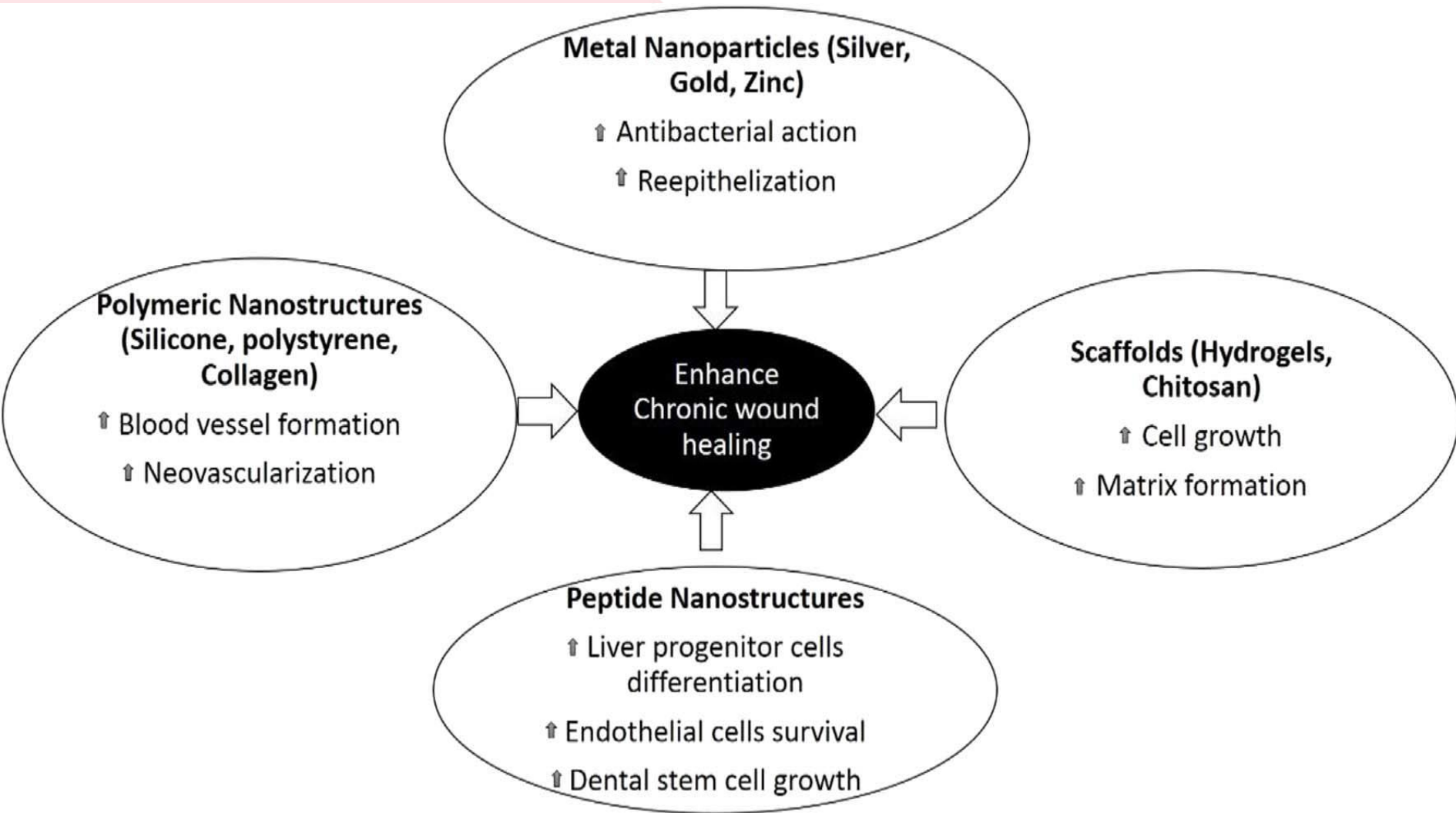


- Fig. 1. Schematic depiction of selected mechanisms of action of bacterial resistance to antibiotic therapy. (Above, left) Reduction of drug accumulation (e.g., down-regulation of porin channels that permit passive inflow of antibiotics or up-regulation of efflux pumps, which actively transport antibiotics out of the cell).
- (Above, center) Modification of antibiotic targets [e.g., modification of penicillin-binding proteins within the cell wall (targets of beta-lactams) or modification of the 30S ribosomal subunit (target of aminoglycosides)].
- (Above, right) Inactivation of antibiotics (e.g., up-regulation of enzyme beta-lactamase which cleaves and inactivates beta-lactams).



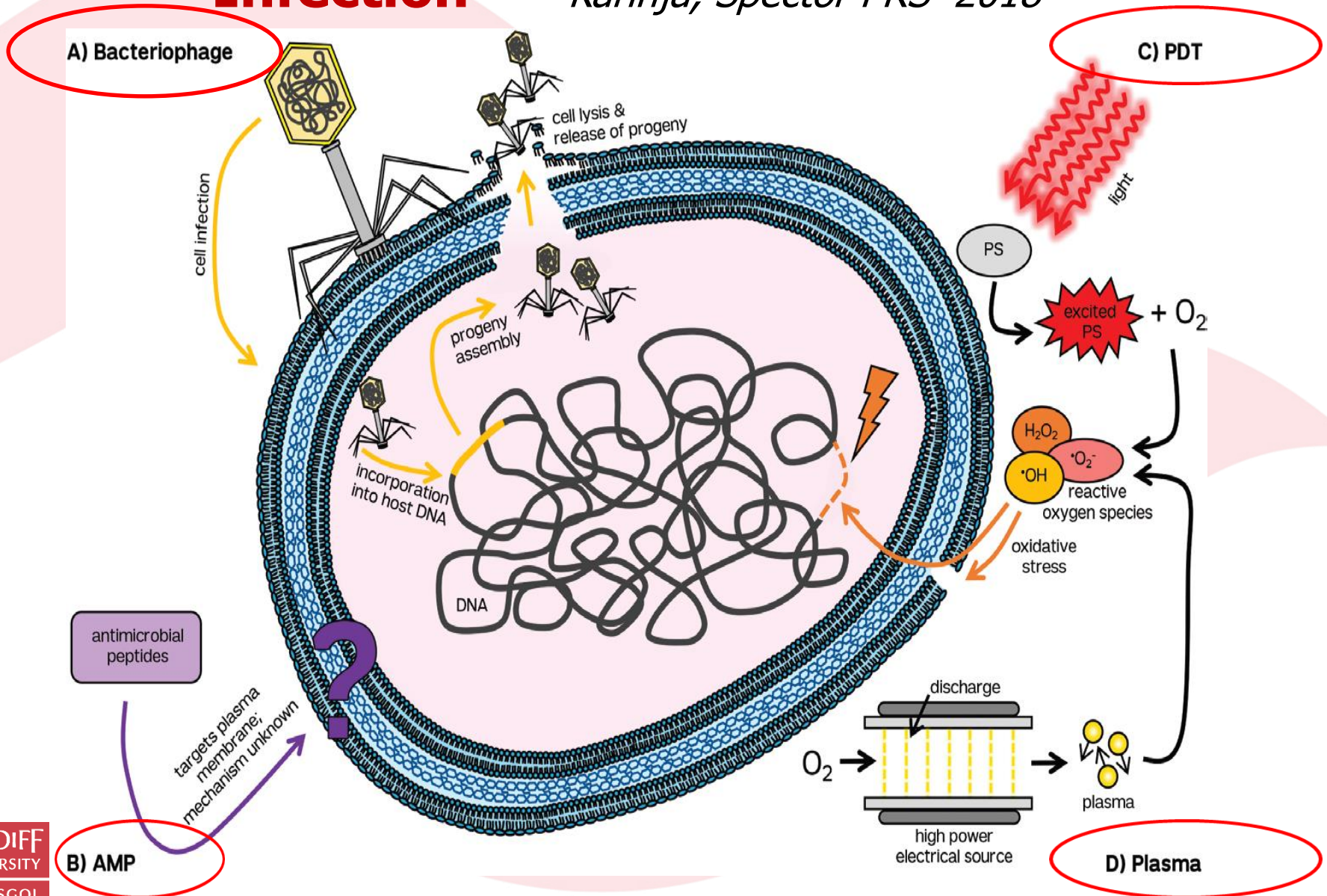
# Potential for Nanoparticles in Wound Healing

*Rajendran et al J Drug Del Sci Tech 2018*



# Rationale for Novel Forms of Treatment of Infection

*Karinja, Spector PRS 2018*



# MicroRNA therapy for infected wounds

**The discovery of microRNAs (miRNAs) in the 1990s led to a revolution in the way scientists think about protein regulation in the cell (Lee et al, 1993). miRNAs are highly conserved, non-coding RNA molecules that average ~22 nucleotides in length. They typically recognize and bind to complementary sequences on messenger RNA (mRNA) molecules, leading to their translational repression or degradation**

A novel approach for identification of functional miRNAs in skin wounds reveals miR-223 as a key regulator of inflammation during tissue repair.

The findings suggest targeting of miR-233 as an effective strategy to enhance neutrophil activation after bacterial infection and to improve healing of infected wounds.

*Hiebert, Werner EMBO Mol Med (2018)*

# Alternatives to Antibiotics

- **Antimicrobial peptides**

produced by microscopic organisms that disrupt bacterial cell membrane

- **Biofilm degrading agents**

Enzymes

pH

High osmolarity

Surfactants

- **QS inhibitors**

In Gm +ve organisms controlled by Auto inducing peptides (AIPs)

In Gm-ve organisms controlled by LuxI and LuxR proteins

Inhibit signal exchange between different bacterial cells

*Buch et al Clin Micro Reviews 2019*

# Alternatives to Antibiotics

- **Misc Compounds**

Gallium compounds influence Iron metabolism in bacteria

Amyloid blockers particularly in E Coli infection

- **Peptidomimetics**

Hinder protein assembly in bacteria

- **Nanoparticles 1-100nm**

Metal NPs

Non metal NPs

Polymeric NPs

Lipid NPs

Quantum dots

Ceramic NPs

- **Scaffolds embedding NPs**

Alginate and Chitosan Hydrogels (DFU)

- **Viral Vector Based Gene Delivery**



# Novel Approaches to Antibiotic Use

Treatment of diabetic foot infections, especially deep-seated ones, remains challenging, in part because impaired blood perfusion and the presence of biofilms can impair the effectiveness of systemic antibiotics. **The local application of antibiotics is an emerging field in the treatment of diabetic foot infections, with demonstrable advantages.** These include delivery of high concentrations of antibiotics in the affected area, limited systemic absorption, and thus negligible side effects. Biodegradable vehicles, such as calcium sulfate beads, are the prototypical system, providing a good elution profile and the ability to be impregnated with a variety of antibiotics, but **the strongest evidence available is for calcium bead implantation for osteomyelitis management**

*Markakis et al IJLEW*

# Downside of Antimicrobial Agents

The use of topical antimicrobials is beneficial for infection control in wound care because wound infection is the major cause of delayed healing. The advantages of topical over systemic antimicrobials include a higher concentration at the target site, fewer systemic adverse effects, and a lower incidence of antimicrobial resistance. Nowadays, topical antimicrobials are divided into three groups: disinfectants, antiseptics, and antibiotics. Only antiseptics and antibiotics can be applied to living skin; therefore, this review will focus only on these groups. The advantages of each topical antimicrobial are well established; however, their disadvantages remain prominent. It is widely known that antiseptics show higher cytotoxicity and a broader spectrum of activity than antibiotics, whereas antibiotics show a higher probability of bacterial resistance development. However, there are still many adverse effects, resulting from each topical antimicrobial.

# Antiseptics can cause problems

*Punjataewakupt 2019*

## **Patient comorbidities/ condition**

**Renal impairment**

**Hepatic impairment**

**Thyroid impairment**

**Respiratory impairment**

**G6PD deficiency**

**Known allergy to sulphonamides**

**Pregnancy and lactation**

-

**Exposed bone or cartilage area**

## **Inadvisable topical antimicrobials**

- Iodine compounds

- Mafenide

- Mupirocin Base

- Neomycin

- Silver sulfadiazine

- All Silver compounds and silver sulfadiazine

- Iodine compounds

- Mafenide

- Mafenide

- Silver sulfadiazine

- Mafenide

Silver sulfadiazine

- Iodine compounds

- Mafenide

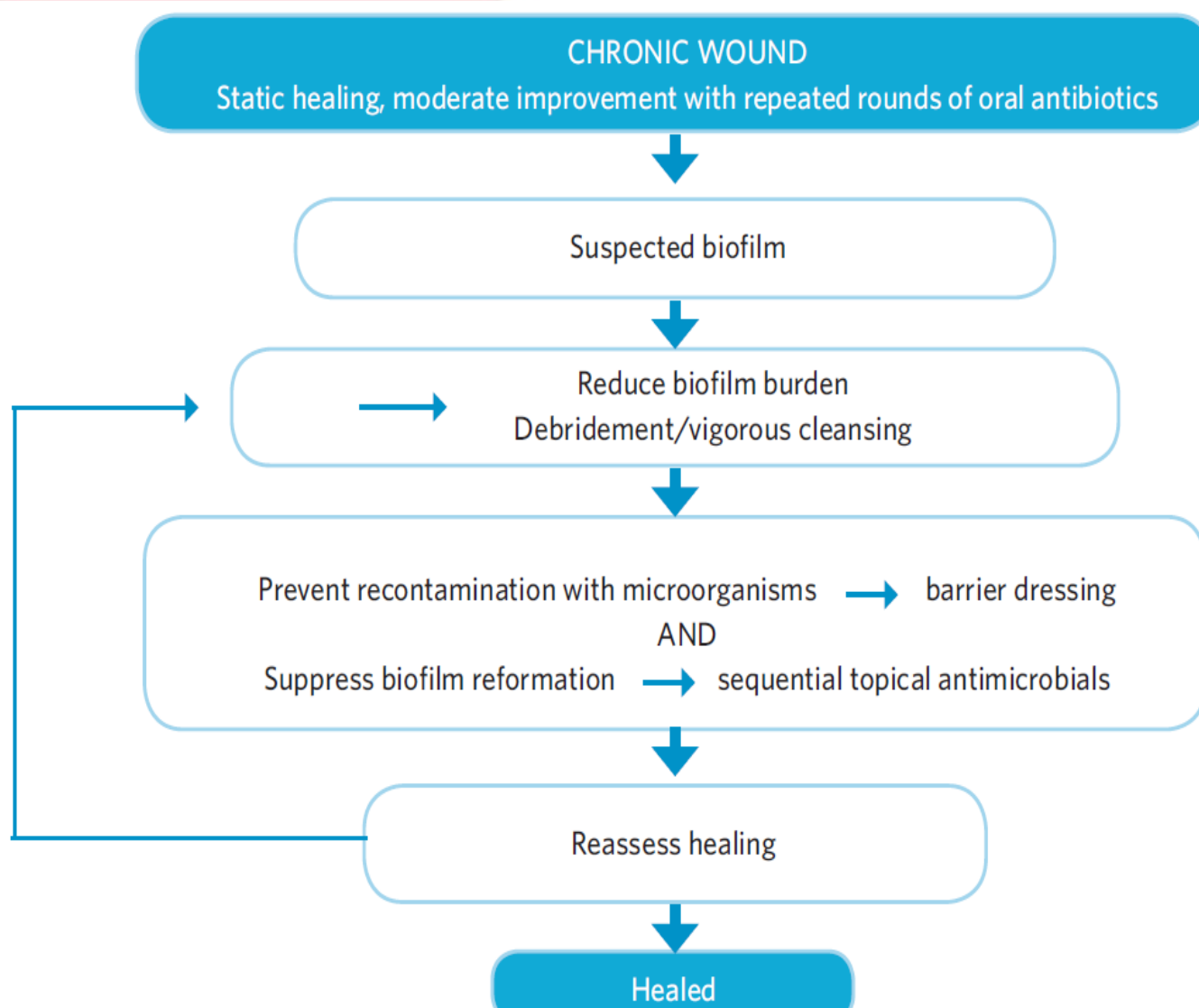
- Mupirocin

- Silver sulfadiazine

- Chlorhexidine

- PHMB

# WUWHS Biofilm Management (2016)



# Consensus on Biofilms in Wounds



- **Wounds that contain biofilms may not be identified**, resulting in ineffective treatment and delayed healing [strong agreement, mean 4.3, SD 0.82].
- **Biofilms are present in most chronic wounds** [strong agreement, mean 4.8, SD 0.42], and are likely to be located both on the surface and in deeper wound layers, but may not be present uniformly across or within the wound [strong agreement mean 4.5, SD 0.97].
- **Wound biofilms are difficult to visualize macroscopically** and slough, debris, and exudate may be visually mistaken for biofilm by clinicians/healthcare professionals [strong agreement, mean 4.6, SD 0.73].
- Important indicators that a **wound is likely to contain a biofilm include recalcitrance to treatment with antibiotics or antiseptics** [strong agreement, mean 4.3, SD 0.67].
- The most important measure for **future diagnostic tests to consider is indication of where the biofilm is located within the wound** [strong agreement, mean 4.0, SD 0.82].



# Consensus on Biofilms in Wounds



- **Debridement is one of the most important treatment strategies against biofilms**, but does not remove all biofilm, and therefore cannot be used alone—this is one of the critical principles of wound bed preparation (tissue, infection/inflammation, moisture balance, and edge of wound) [strong agreement, mean 4.9, SD 0.32].
- **Biofilms can reform rapidly**; repeated debridement alone is unlikely to prevent biofilm regrowth; however, effective topical antiseptic application within this time-dependent window can suppress biofilm reformation [strong agreement, mean 4.0, SD 0.67].
- **Topical antiseptics that are effective antibiofilm treatments should have strong antibiofilm effects in appropriate in vitro test models against mature biofilms** [strong agreement, mean 4.0, SD 0.67].
- In vitro biofilm methods with clinically relevant test conditions are useful to screen treatments for their antibiofilm efficacy [strong agreement, mean 4.5, SD 0.71].
- RCTs and comparative clinical evidence of antibiofilm treatment should be used to support clinical guidelines, protocols, and treatment choices. However, **in the absence of RCT-level data, antibiofilm interventions should be supported by RCT evidence of the broader impact on wound healing** [strong agreement, mean 4.2, SD 0.79].

*Schultz et al. Wound Rep Reg (2017) 25 744–757 WHS*

# Do not get despondent about wound infection !



# Resources



- <http://www.woundsinternational.com/consensus-documents/view/iwii-wound-infection-in-clinical-practice>
- <http://www.woundsinternational.com/wuwhs/view/position-document-management-of-biofilm>
- [http://www.wounds-uk.com/pdf/content\\_11958.pdf](http://www.wounds-uk.com/pdf/content_11958.pdf)
- [http://www.wounds-uk.com/pdf/content\\_10964.pdf](http://www.wounds-uk.com/pdf/content_10964.pdf)

# Webcast resources



- <http://www.woundsinternational.com/videos/view/understanding-biofilm-based-wound-care-what-you-need-to-know>
- <http://www.woundsinternational.com/videos/view/microbiology-an-overview-of-current-issues-including-resistance-and-biofilms>





# Thank You!

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