Wound Infections and Microbiology – The future

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Wound ?

Structural damage of (skin)tissue, by any injury, results in a wound:

- open wound: cut, punctured
- closed wound: contusion, hematoma

Management and Complications

- Management: function of wound type
  - Cleaning , debridement, closure, dressings and **antibiotics…if microbial contamination** (positive cultures) and **inflammation**
- Complications:
  - Infection can impede healing and evolve into life threatening situations
  - Global wound care13-15 $ billion annually

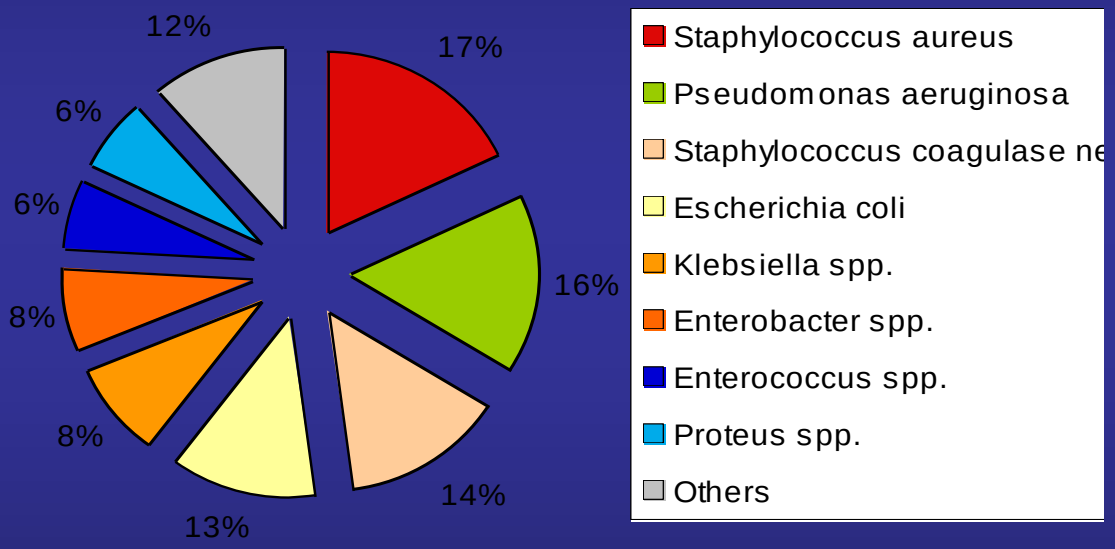
Colonization ▸Infection = continuum!

Siddiqui and Bernstein, Clinics in Dermatology, 2010.

Infection

- **Infection is the detrimental colonization of a host, or host site, by micro-organisms (pathogenic/opportunistic)**
- **It is an interplay between the host and the microbes where immune status, microbial load and wound extension are major parameters.**
- **Wound-Infection is comparable with an ‘ecological catastrophe’ where opportunists and/or pathogens ‘fill the niche’**

Pathogens in Burns



Pirnay *et.al., J.Clin. Microbiol*. 2003.

Staphylococcus aureus remains common cause early burn wound infection

Pseudomonas aeruginosa from the patient's endogenous gastrointestinal flora and/or an environmental source is the **most common cause of burn wound infections in many centers.**

Altoparlak *et al*., *Burns* 2004.
Church *et al*., *Clin. Microbiol. Rev*. 2006

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Antibiotic Resistance

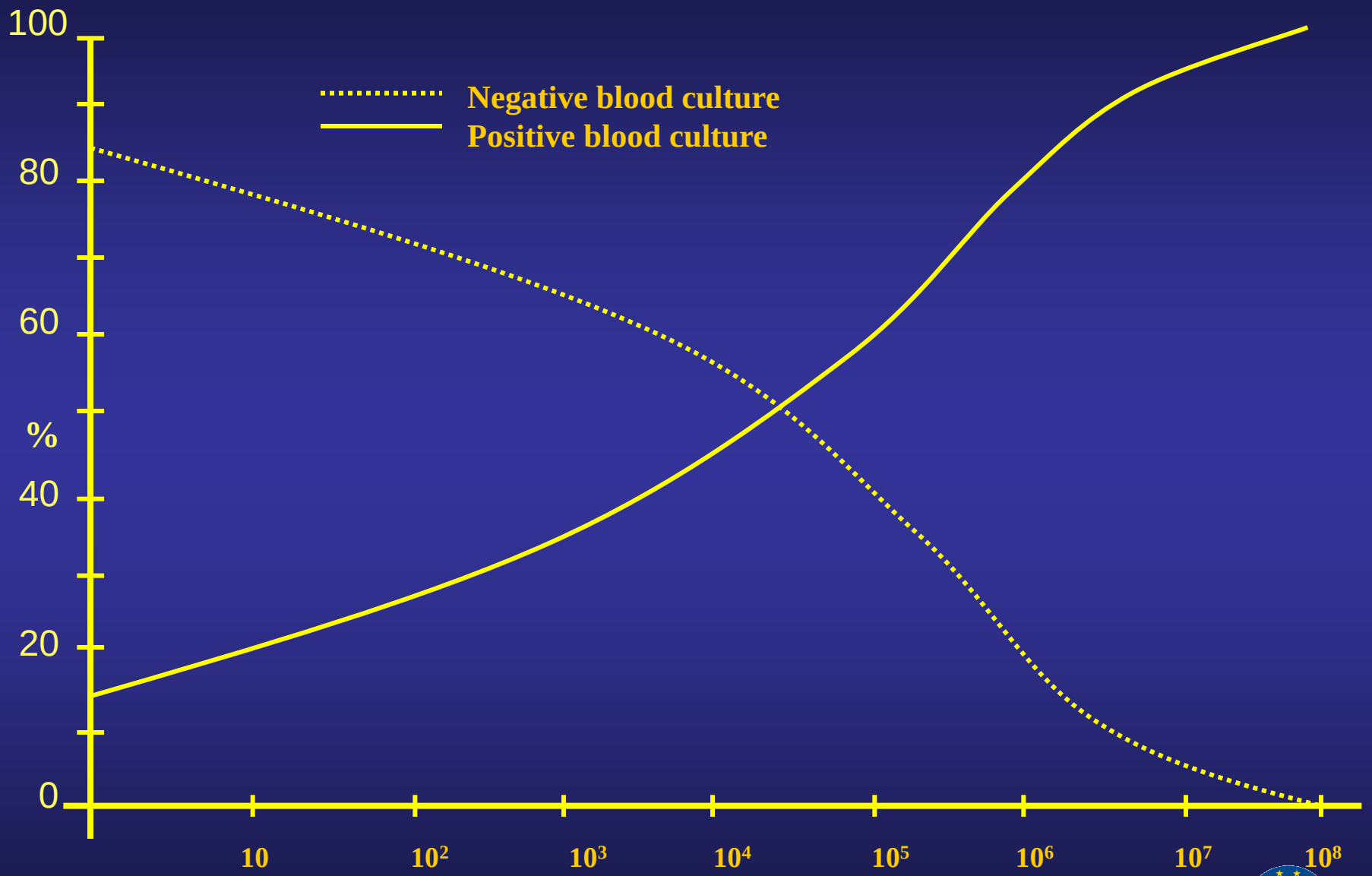
- What happened to antibiotics? Once considered the universal answer to infectious disease, we now know the effective life span of these once-miraculous drugs is limited. The problem, simply, is that we "got complacent" (Barry Kreiswirth)

- Bad understanding of evolutionary biology (mechanistic medicine) $\rightarrow$ use of non-evolving anti-microbials $\rightarrow$ resistance

Possible answers/Solutions?

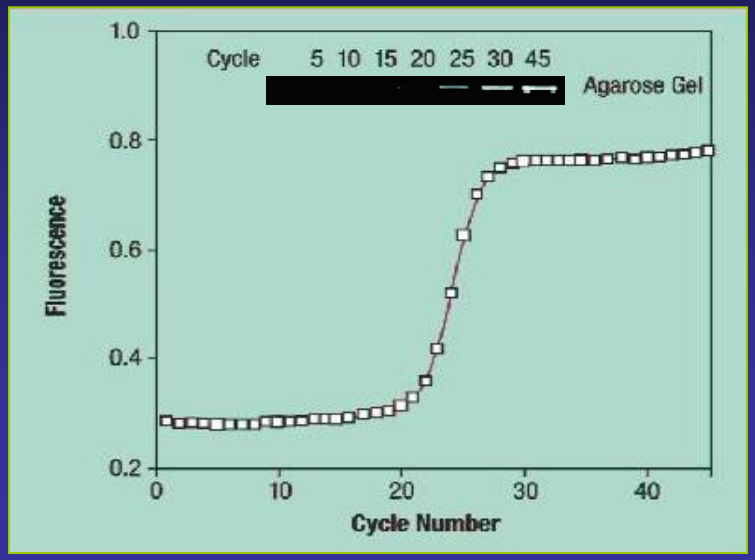
- New antibiotics (AB)
- Better use of AB
- New rapid molecular diagnostics
- Interplay with host's immune system
- Vaccines
- New anti-microbial approaches based on "old" traditions and new basic biology insights (evolution and host-microbial interactions)

Relation between cutaneous bacterial count and passage of the germ into the blood

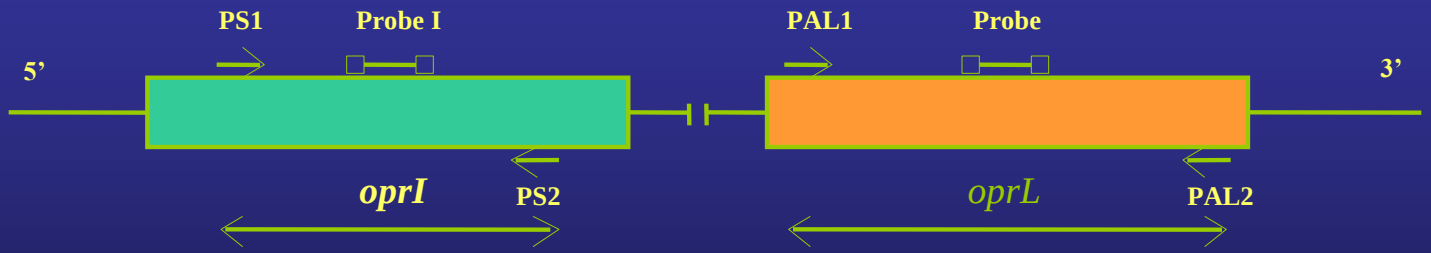


Perez-Cappelano et al., 1976 , Burns Vol 3, 42-45

Quantitation of *Pseudomonas aeruginosa* in wound biopsy samples: from bacterial culture to rapid 'real-time' polymerase chain reaction (Pirnay *et al*., Crit Care,4, 255-261, 2000. Jannes and De Vos, Diagnostic bacteriology protocols, 2006)



Amplification of the lipoprotein genes *oprI* and *oprL*



Turn around time from sample collection to result, in less then 1 hour

‘New’ or ‘re-discovered’ approaches

- Quorum sensing and biofilm interactions
- Use of other organisms or biological entities as anti microbial (pathogenic) agents: maggots and bacteriophages

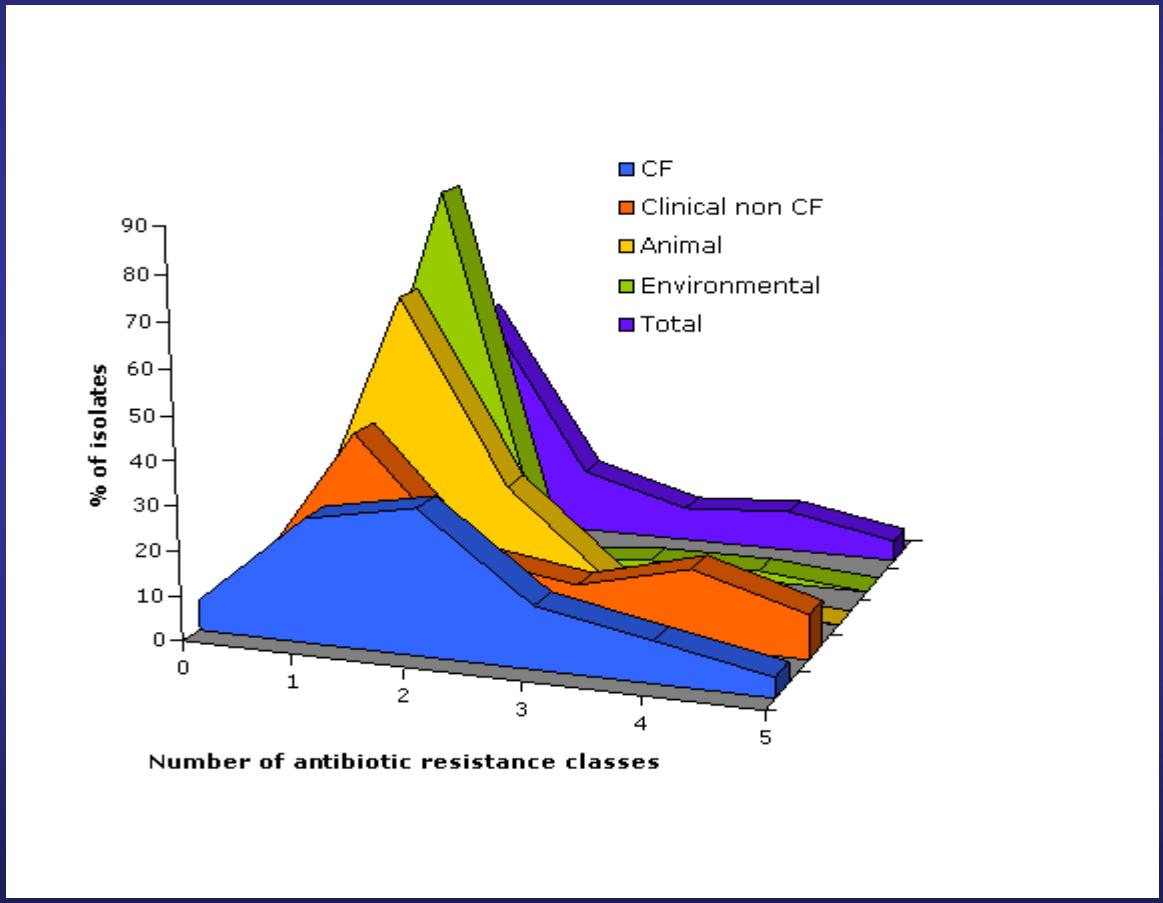
Quorum sensing (QS)

- Bacterial life modus: **planktonic or biofilm**
- QS: **bacterial communication system**
  - ❑- Mediated by small difusible molecules
    - **oligopeptides in Gram+**
    - **N-acylhomoserine lactones,AHLs,in Gram -**
    - **auto-inducers,AI, in G+/-**
  - ❑- At a critical concentration, cell density dependent, the bacterial gene expression alternates ▸virulence factors, biofilm…
  - ❑- QS occurs within a single species as well as between different species. (complexity of wound microbiology)
- Potential anti infectious agents: **QS inhibitors / quenchers:** garlic, *Delicea pulchra* red sea- weed…fungal extracts (Bjarnsholt T, *Nature Protocols,* 2010; Rasmussen *J.Bact*, 2005)

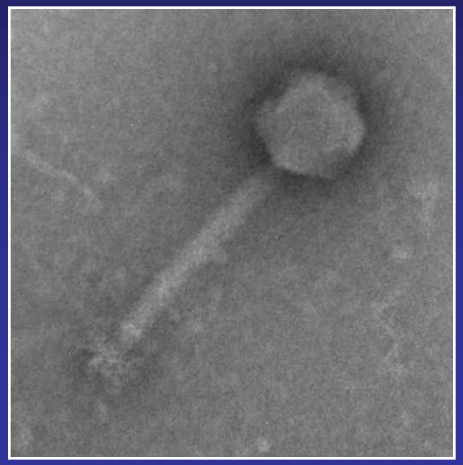
Biofilm (BF)

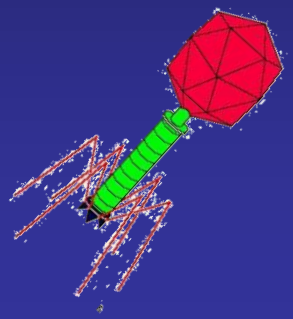
- **A complex aggregation of bacteria embedded in a self produced extracellular polymeric matrix**
  - Bacterial biofilm lifestyle ▸ different gene expression…**influence on AB-R!**
  - US NIH estimates **> 80% infections BF**
  - **Impairs wound healing** and **reduce topical efficiency**
  - Biofilm inhibitors ▸ direct interactions with chemicals or interaction with foreign organismal signaling molecules

Different antibiotic mechanisms in *P.aeruginosa* isolates from different origin



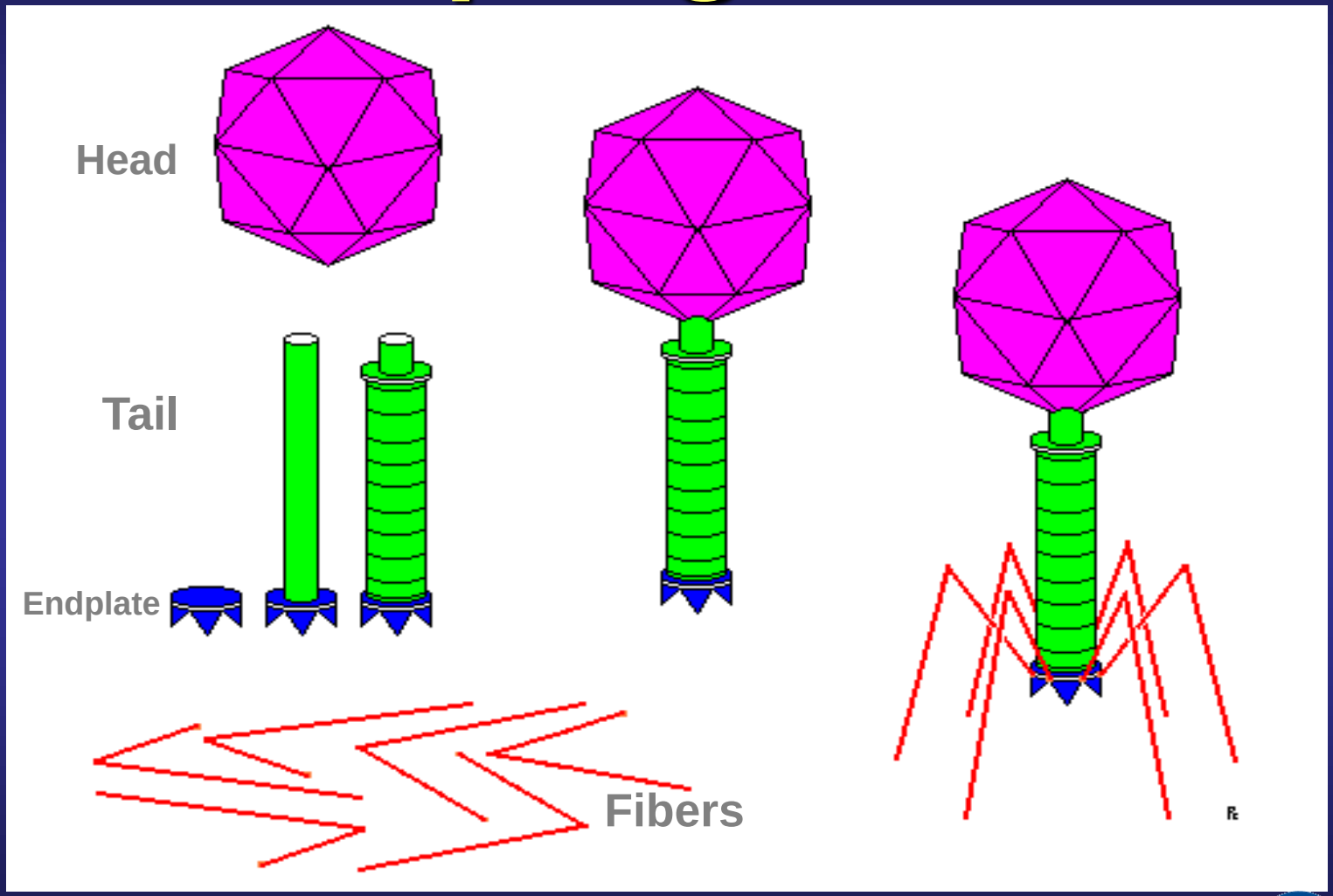
Bacteriophages: natural biological entities





- The most abundant natural lifelike biological entity in our biosphere (>10^9/ml sea water, role in origin and evolution life)
- Evolving anti-microbial (“Darwinian Medicine”)
- ‘Viruses’ of bacteria (including “Superbugs”)
- Co-evolving with bacteria
- Natural enemies/controllers of bacteria

Bacteriophage structure



Phage Therapy ?

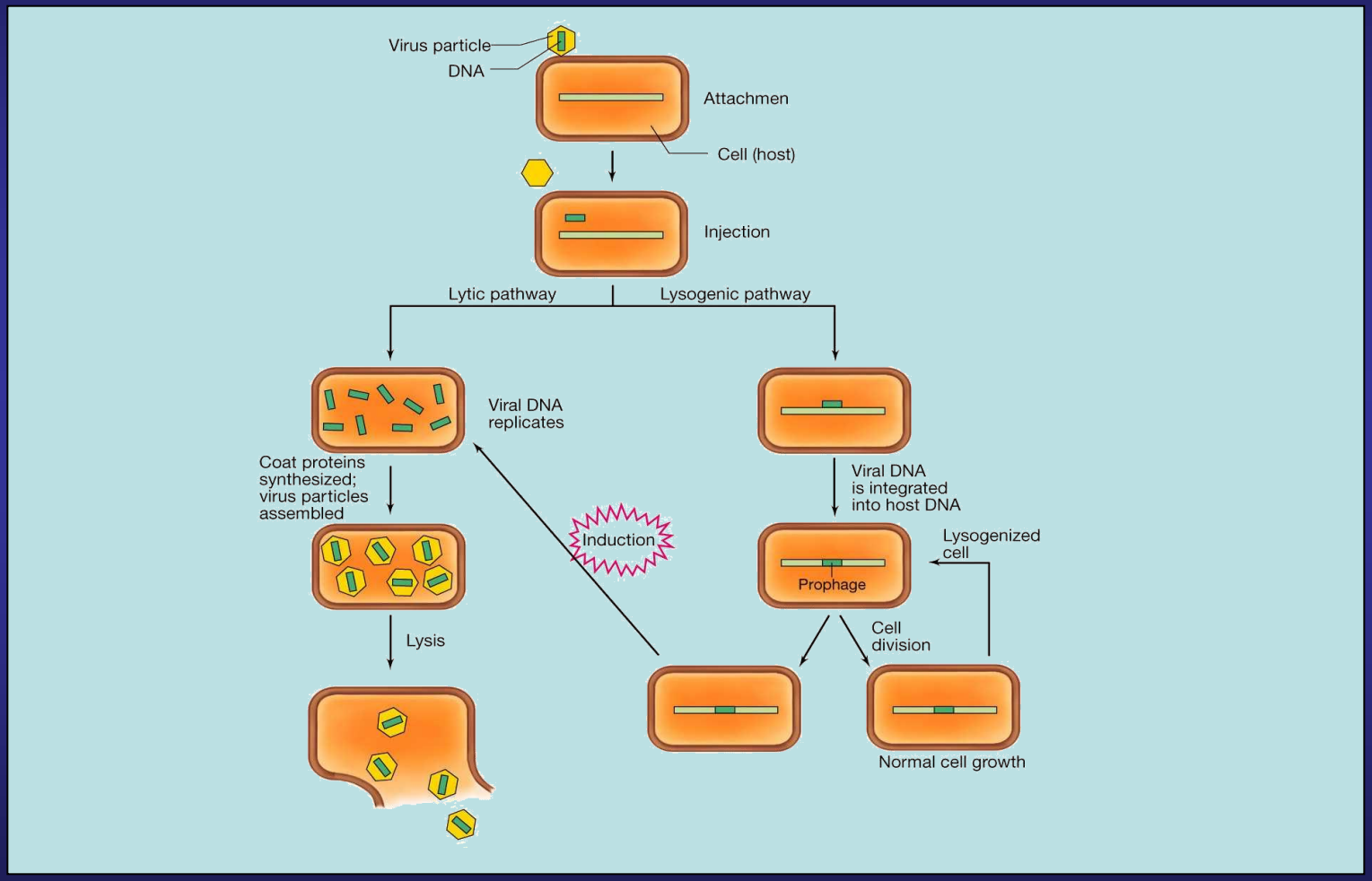
a potential **Alternative** to the **World Wide Antibiotic Resistance** problematic?

Bacteriophage therapy consists in the application of bacteriophages (“bacteria eaters”, natural ennemies of bacteria) for killing noxious bacteria

Still no eucaryotic infections with phages have been reported, nor were any phage sequences detected in the human genome

Natural controllers and co-evolutive

How Phages Kill Bacteria



Phages vs. Antibiotics

PHAGE

- Very specific (e.g. part of the strains of one species)
- Development of new phage preparations: relatively easy and inexpensive
- Disease causing agents has to be known *(for now)*
- No known side effects

ANTIBIOTIC

- Non specific (including commensal flora)
- Development of new antibiotics: time-consuming and expensive
- Broad spectrum, agents can be unknown
- Multiple side effects

East vs. West

Frederic Twort
UK 1915

Felix d'Hérelle
France/Canada 1917

"User-friendly" ANTIBIOTICS
1940s - Today

Antibiotic resistance
BWC/QAMH

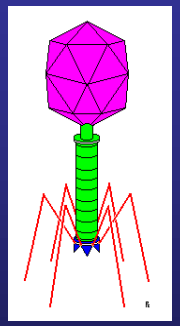
WEST

Stalin WWII

EAST

George Eliava
Georgia, USSR

Eliava Institute, Tbilisi,
Georgia 1923 - Today

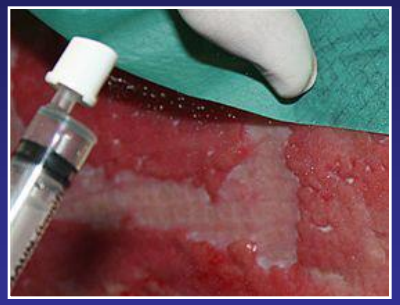


Phages

Dr. Maya Merabishvili

Therapeutic Phage Application on Burns

A safety study



- The modest and very cautious protocol did not allow an adequate evaluation of efficacy
- The application of BFC-1 elicited no adverse events whatsoever
- The medical and nursing staff grew familiar with phages and deem them safe for topical use on infected burn wounds
- This study, represents an essential and necessary step towards the eventual acceptance of phage therapy

Verbeken *et al*., *Future Medicine*,2, 485-491, 2007.

Merabishvili *et al*., *PLoS ONE.* 2009; 4(3): e4944.

Kutter *et al, Current Pharmaceutical Biotechnology,11, 69-86, 2010.*

Phage application on a shot wound in Gori Military Hospital, Georgia

Phage Application at the Queen Astrid Military Hospital, Brussels, Belgium

Conclusion

- Scientific insights and technological developments show promising avenues. ***But!***
- Subjective feelings like **'seeing viruses as enemies of life'** or **'maggots as the seeds of rottening'** hamper positive progress ***as well as*** **isssues** related to the **regulatory affairs** and **patents** (based on 'classic mechanical' worldvieuws).
- **But**, inherent naturally, **we will have to adapt!**
- Thus: **Interdisciplinary "*interactions and research*"** is the message**!**

Acknowledgement

- Dr. Jean-Paul Pirnay, Director of the LabMCT
- Dr. Serge Jennes, Head of the Burn Wound Centre (BWC)
- Dr. Thomas Rose, Surgeon at the BWC and Director of the Keratinocytes and Skin banks of the BWC
- Mr. Gilbert Verbeken, biologist and QA/QC manager of the Keratinocytes and Skin banks of the BWC
- Dr. Patrick Soentjens, Infectiologist of the BWC

- Collaborative groups:

- Prof. Mario Vaneechoutte, UGent, Belgium
- Dr. Maya Merabishvili from the Eliava Institute , Tbilisi, Georgia
- Prof. Rob Lavigne and Prof. Isabelle Huys of KULeuven, Belgium
- Dr. Laurent Debarbieux, Pasteur Institute, Paris, France
- Prof. Elisabeth Kutter, Evergreen State College, Washington, USA