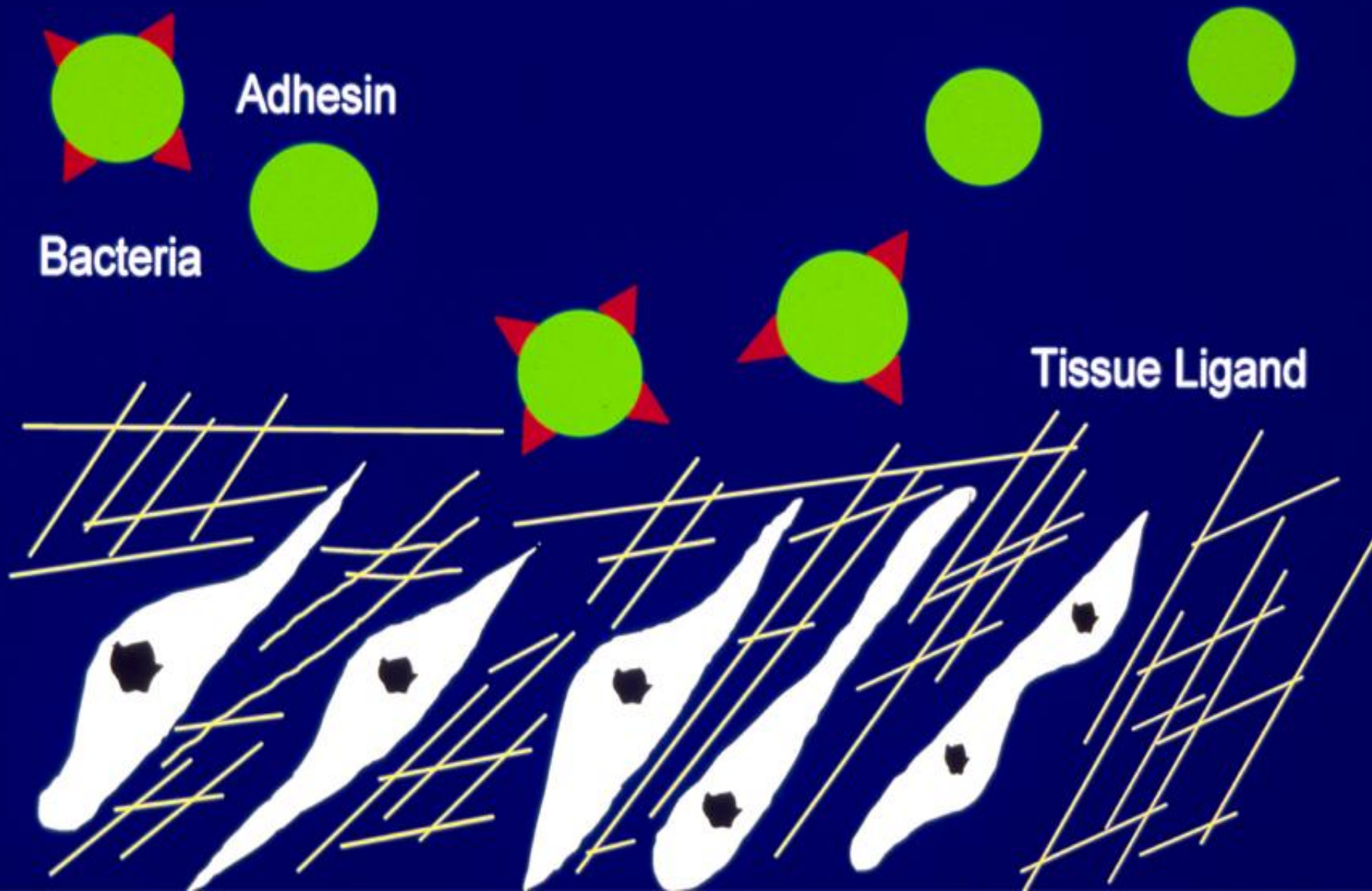


INFECTIONS ARE CAUSED BY BACTERIA ADHERING TO HOST TISSUE



“MSCRAMMS”

Microbial

Surface

Components

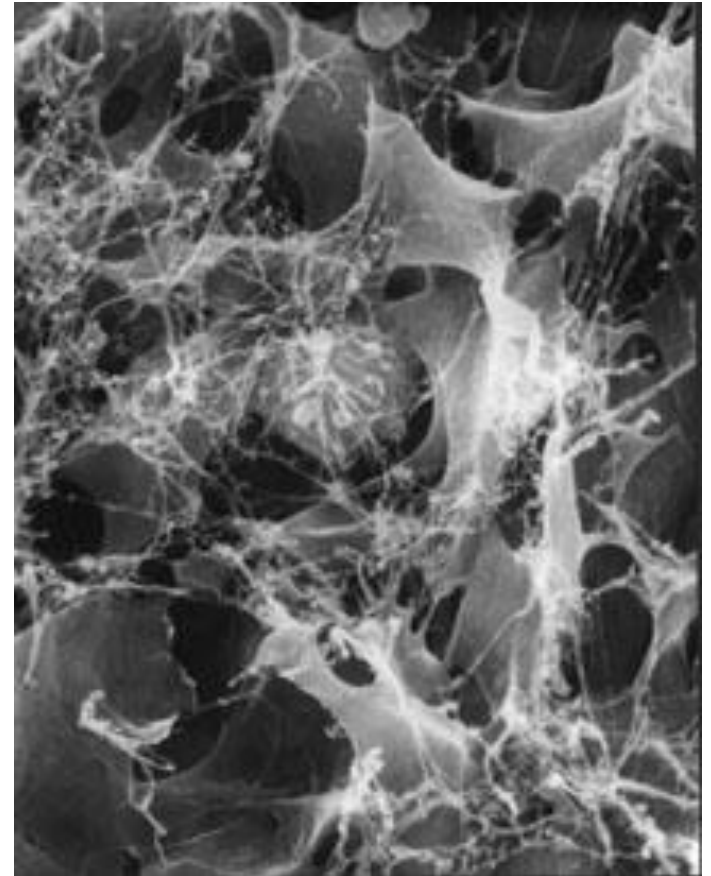
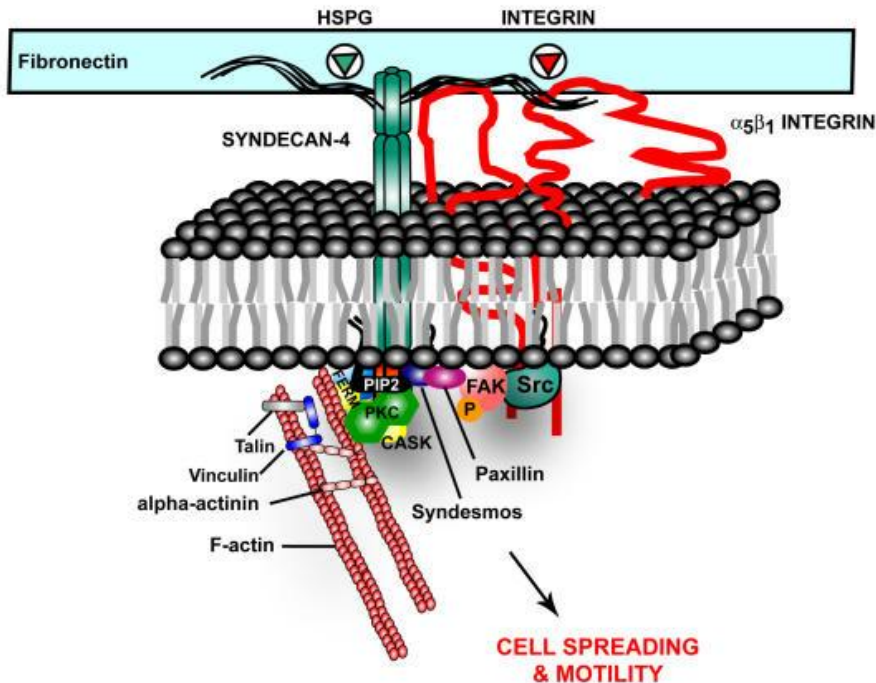
Recognizing

Adhesive

Matrix

Molecules

What is Matrix?



MSCRAMMs

| Microorganism | MSCRAMM | Ligand |
|------------------------|----------------------------------|------------------------|
| <i>B. burgdorferi</i> | DbpA, DbpB | Decorin |
| <i>T. pallidum</i> | P1, P2 | Fibronectin |
| <i>T. denticola</i> | Msp | Laminin |
| <i>S. aureus</i> | FnbpA, FnbpB, ClfA, ClfB, CNA | Fibrinogen/Fibronectin |
| <i>S. dysgalactiae</i> | S2 | Fibronectin |
| <i>S. pyogenes</i> | SfbI, SfbII | Fibronectin |
| <i>M. leprae</i> | FAP | Fibronectin |







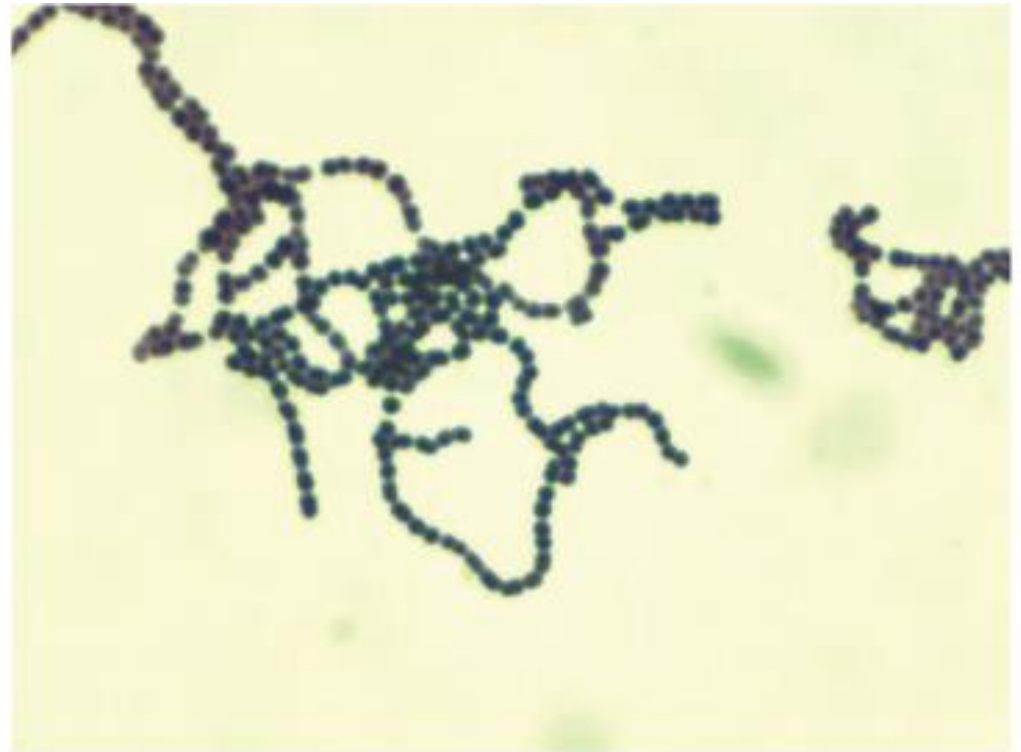
Streptococcus & Enterococcus

'He prayeth best who loveth best
All things both great and small.
The Streptococcus is the test--
I love it least of all.'

--Wallace Wilson

General Characteristics

- **Gram-positive spherical cells.**
 - appear more elongated than spherical
- **Arranged in chains or pairs.**
 - more likely to form chains in liquid culture.
- **Metabolism fermentive.**
 - Lactic acid primary product
- **Catalase negative**
- **Most are facultative anaerobes.**

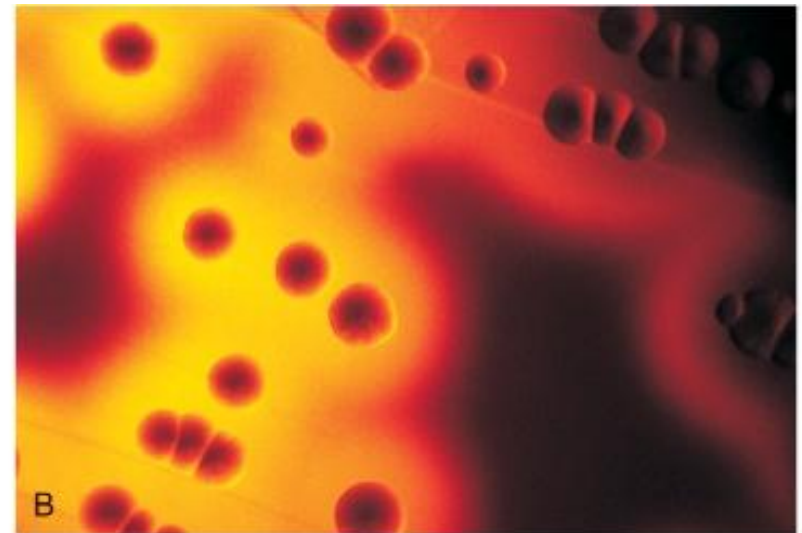
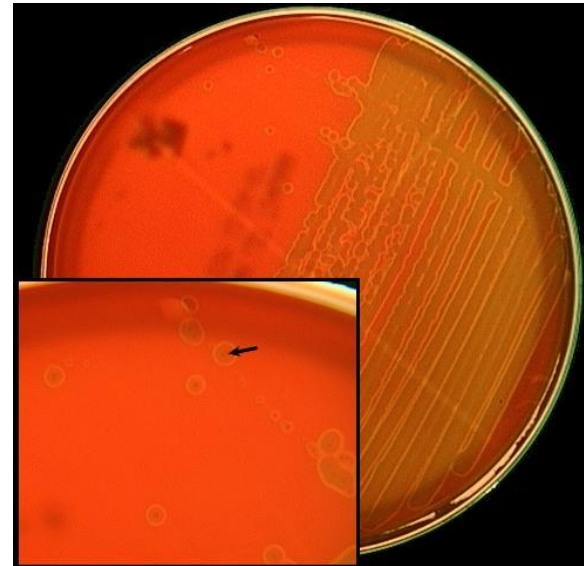


Classification

- Hemolytic pattern on sheep's blood agar.
- Physiologic characteristics (type of infection , site of origin).
- Serologic group or type of C carbohydrate (Lancefield classification).
- Biochemical characteristics.

General Characteristics-Types of Hemolysis

- **Alpha (α)-partial lysis of RBCs around colony.**
 - greenish discoloration of area around colony.
- **Beta (β)-complete RBC lysis.**
 - clear area around colony
- **Nonhemolytic-no lysis of RBCs.**
 - no change in agar
- **Alpha-prime (α')-Small area of intact RBCs around colony surrounded by a wider zone of complete hemolysis.**



General Characteristics - Types of Hemolysis

| Hemolysis | Description |
|---------------------------|---|
| Alpha (α) | Partial lysis of RBCs around colony; greenish discoloration. |
| Beta (β) | Complete lysis of RBCs around colony. |
| Nonhemolytic | No lysis of RBCs; no change in color. |
| Alpha-prime (α') | Small area of intact RBCs around colony surrounded by a wider zone of complete hemolysis. |

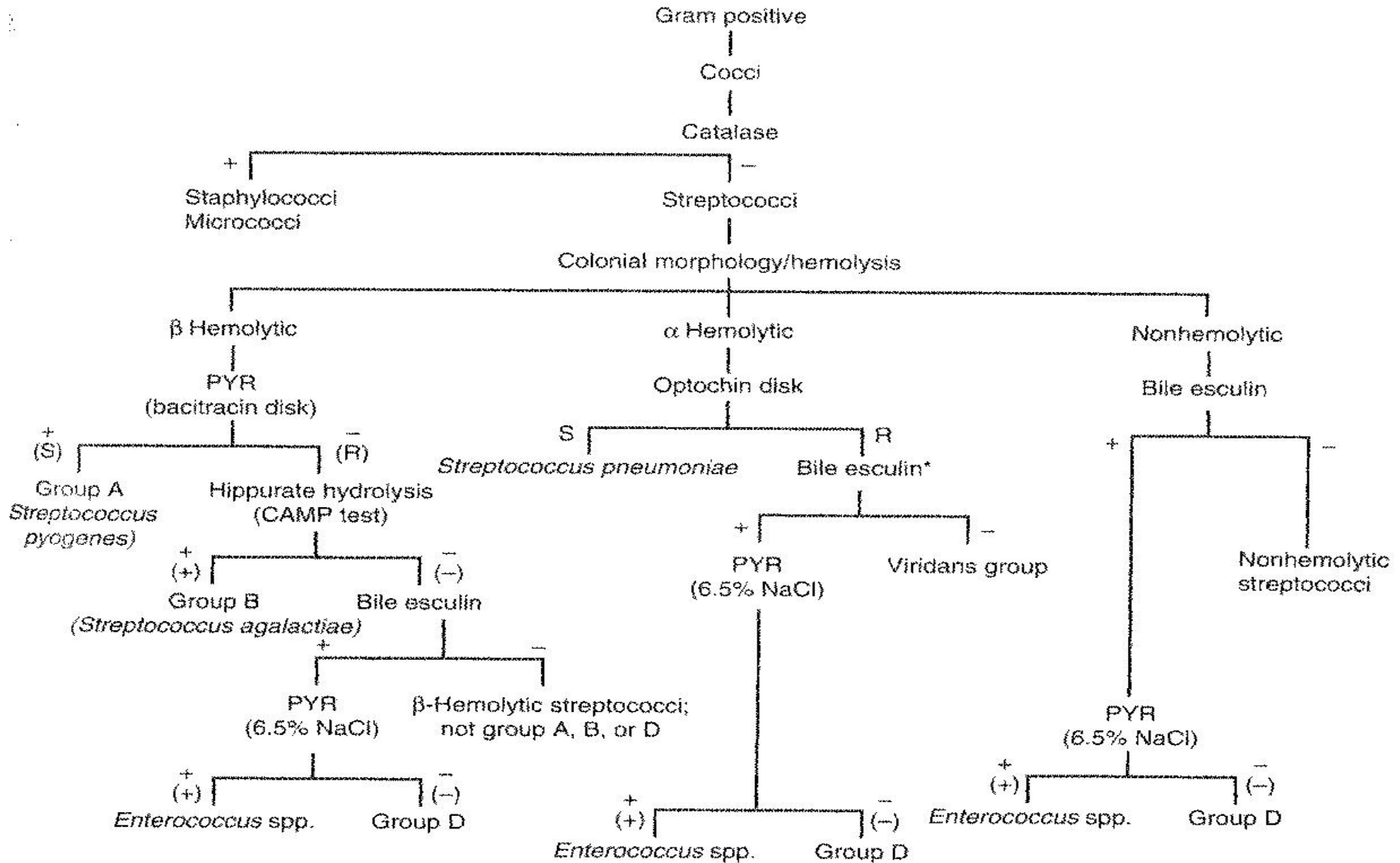
Classification

- Lancefield classification scheme
 - dependent upon extraction of **C carbohydrates** by placing the organism in dilute acid and heating for 10 min.
 - Lancefield was able to divide the streptococci into serologic groups (designated by letters A-F).
- Biochemical identification
 - Susceptibility testing
 - **Bacitracin** has been used for years to differentiate group A streptococci from other groups of β -hemolytic streptococci.
 - Low concentrations (0.04U) **selectively** inhibits the growth of group A streptococci.
 - **CONVERSELY**, groups A and B are resistant to sulfamethoxazole and trimethoprim (STX), whereas other groups are sensitive.

Classification of Streptococcus and Enterococcus

| Species | Antigen Type/ Hemolysin type | Common Name | Disease |
|---|---------------------------------|-----------------------|--|
| <i>S. pyogenes</i> | A/ β * | Group A Strep | Rheumatic fever, scarlet fever, pharyngitis, glomerulonephritis, pyogenic infections |
| <i>S. agalactiae</i> | B/ β * | Group B Strep | Neonatal sepsis, meningitis, puerperal fever, pyogenic infections |
| <i>S. equisimilis</i> <i>S. equi sp. zooepidemicus</i> <i>S. equi</i> | C/ β | Group C Strep | Pharyngitis(?), impetigo, pyogenic infections |
| <i>S. bovis</i> <i>S. equinus</i> | D/ α * | Nonenterococcus | Endocarditis, UTI, pyogenic infections |
| <i>E. faecalis</i> <i>E. faecium</i> <i>E. durans</i> | D/ α, β * | Enterococcus | UTI, pyogenic infections |
| <i>S. pneumoniae</i> | -/ α | Pneumococcus | Pneumonia, meningitis, pyogenic infection |
| <i>S. mutans, sanguis, mitis, salivarius</i> | -/ α | Viridans strep | Endocarditis, dental caries, abscesses in various tissues |

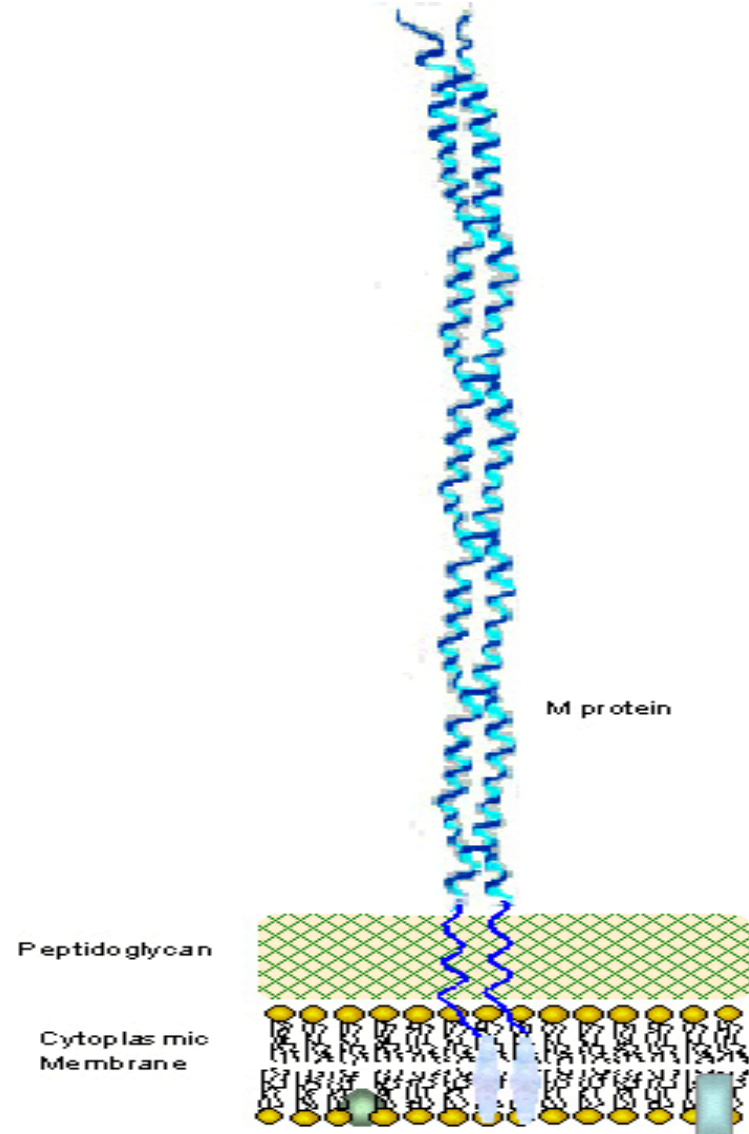
Schematic for the presumptive identification of Gram+ cocci



*Perform additional tests if isolate is from nonrespiratory source.

Group A Virulence Factors

- M protein
 - only found on Group A streptococcus
 - **essential to virulence**
 - resist phagocytosis
 - attach to mucosal cells
- More than 80 different serotypes.
- Protection against infections is related to the presence of type-specific anti-M antibodies.



About 9,000-11,500 cases of invasive GAS disease occur each year in the United States, resulting in 1,000-1,800 deaths annually. STSS and necrotizing fasciitis each comprise an average of about 6%-7% of these invasive cases. In contrast, there are several million cases of strep throat and impetigo each year.

Group A Virulence Factors

- Hyaluronidase (spreading factor)
 - bacterial spread
- Erythrogenic toxin (exotoxin)
 - A, B, and C.
 - Responsible for the red spreading rash referred to as 'scarlet fever'.
 - Role, if any, in pathogenesis is not known.
- Streptolysin S
 - oxygen stable
 - non antigenic
 - also lyses leukocytes
- Streptolysin O
 - Oxygen labile.
 - needs to be in an anaerobic environment to work
 - Lysis leukocytes, platelets, and other tissue cells as well as RBCs.
 - highly antigenic
 - used for diagnostic purposes

Group A Infections

- Pharyngitis

- most common clinical expression of GAS
 - majority of cases of bacterial pharyngitis are due to GAS, although both Groups C and G can manifest this way too.
- Strep throat is most commonly seen in children between 5-15 years of age
 - sore throat, malaise, fever, headache, vomiting and nausea. Nausea, vomiting and abdominal pain can occur.
 - **1/3 of those complaining of a sore throat have culture positive of *S. pyogenes*.**

- Pyodermal Infections

- Impetigo
- Erysipelas →
 - infection of skin and subcutaneous tissues.
 - it is a spreading lesion that is rare.
- Cellulitis
- Scarlet fever
 - 1-2 days after infection--last 5-7 days
 - Rash that appears on the upper chest and spreads to the trunk and extremities.



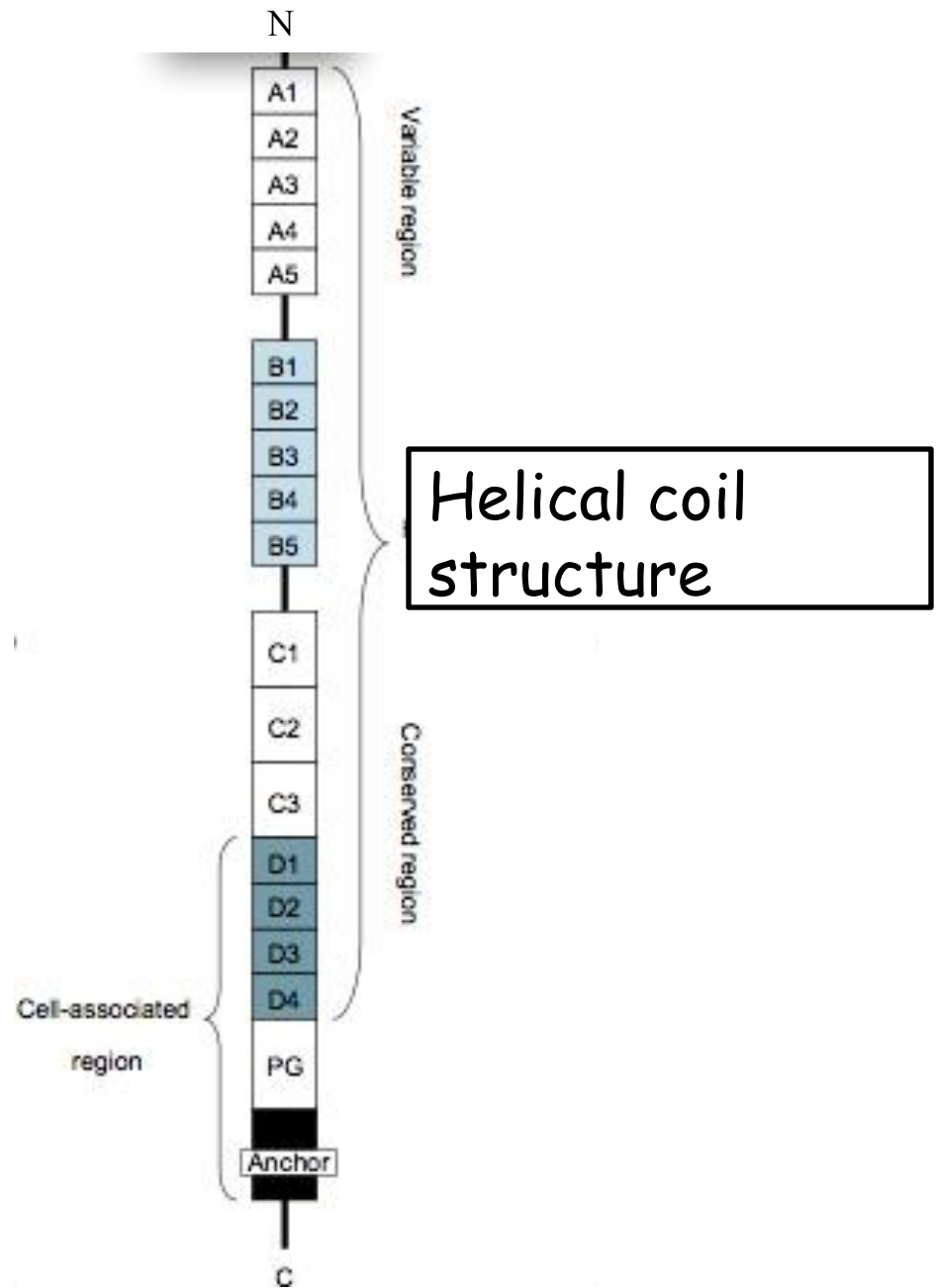
Group A--Poststreptococcal Infection

- Rheumatic fever
 - complication of GAS pharyngitis
 - characterized by fever
 - inflammation of heart, joints, subcutaneous tissue, and blood vessels
 - attacks begin around 1 month post infection
 - chronic, progressive damage to heart valves
 - cross reactivity to heart tissue
 - direct infection of the heart tissue
 - direct toxicity resulting from bacterial exotoxins
- Glomerulonephritis
 - subsequent to either pharyngitis or cutaneous infection
 - more common in children
 - circulating immune complexes

Penicillin/Erythromycin drugs of choice for treatment.

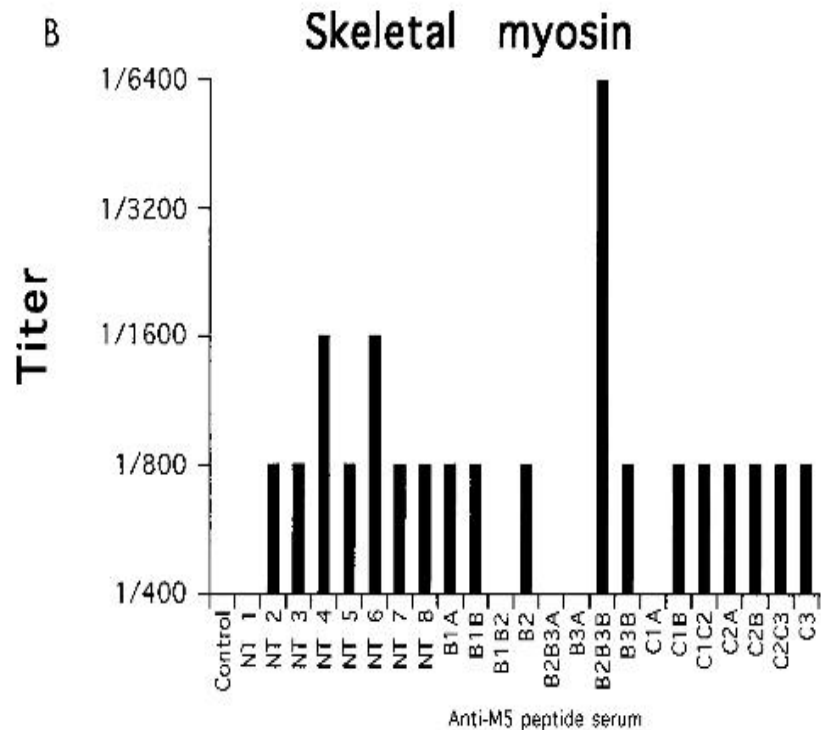
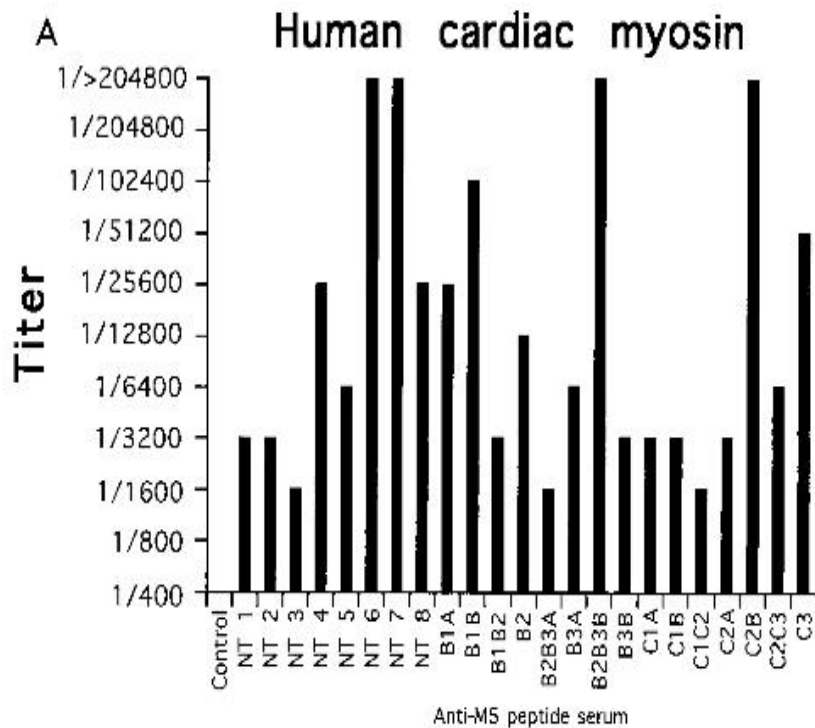
Olive et al. Expert
Rev. Vaccines. 2004.
3:43-58

Diagrammatic
representatio
n of the GAS
M protein
illustrating
the different
repeat
regions and
degrees of
diversity.



Reaction of anti-M5 peptide sera with human cardiac myosin by ELISA

- Cunningham et al. Infect. Immun. 1997. 65:3913-3923



Group A--Invasive Infections

- **Stretptococcal toxic shock syndrome**
- **Endocarditis**
 - Group A and B are the most common cause of endocarditis
- **Necrotizing fasciitis**
 - 'necrotizing erysipelas'
 - 'hospital gangrene'
 - primarily *GAS*
 - some staphylococci and clostridial species can cause similar lesions



Group B--Virulence Factors and Disease

- Capsule primary defense mechanism
 - prevents phagocytosis but is ineffective after opsonization.
- Disease
 - GBS known initially for causing mastitis in cattle.
 - Significant cause of invasive disease in the newborn
 - infection associated with obstetric complications.
 - recommended that all pregnant women be screened for GBS at 35-37 weeks gestation.

Group C, D and G-Virulence Factors and Disease

- Despite their different Lancefield antigens groups C and G are now believed to belong to the same species.
 - The name *S. dysgalactiae* has been proposed for strains isolated from human infections.
- Group D-
 - Group D (*S. bovis*, *S. equinus*)
 - Enterococcus



S. pneumoniae

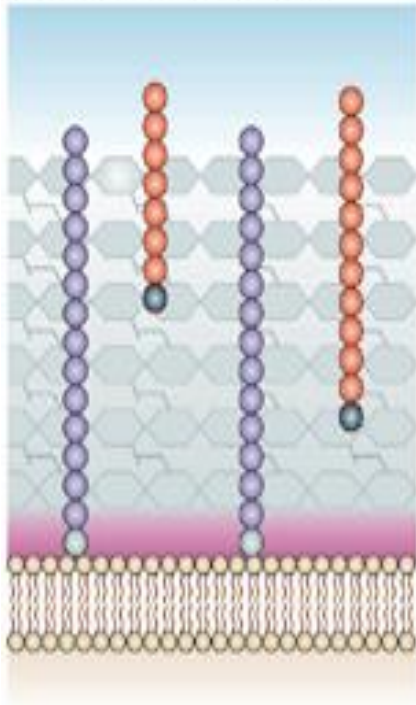
- *Encapsulated, Gram-positive coccus.*
- *Oval, arranged in pairs or short chains.*
- *Fastidious nutritional requirements.*
- *The capsular polysaccharides have been used for the serologic classification of strains; currently **90 serotypes** recognized.*

S. pneumoniae-Cell Wall

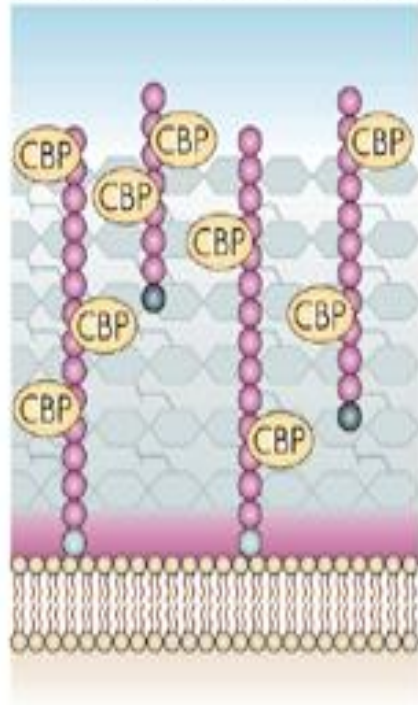
- Like all Gram-positive organisms contains a peptidoglycan layer of the cell wall.
- However, the teichoic acid associated with the cell wall is also rich in **choline** which is unique to the cell wall of *S. pneumoniae*.
- Two forms of teichoic acid exist:
 - One exposed on the cell surface
 - This species specific structure is called the **C-polysaccharide**. Linked to the peptidoglycan.
 - Not related to the group-specific carbohydrate observed by Lancefield.
 - The other form of teichoic acid covalently bound to the plasma membrane lipids.
 - This form also called F antigen because it cross-reacts with the Forssman surface antigens on mammalian cells.

Cell wall glycopolymers (CWGs) in the cell walls of major human pathogens

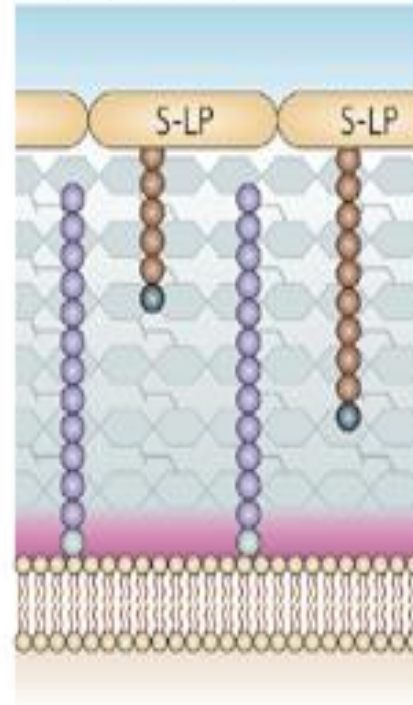
Staphylococcus aureus



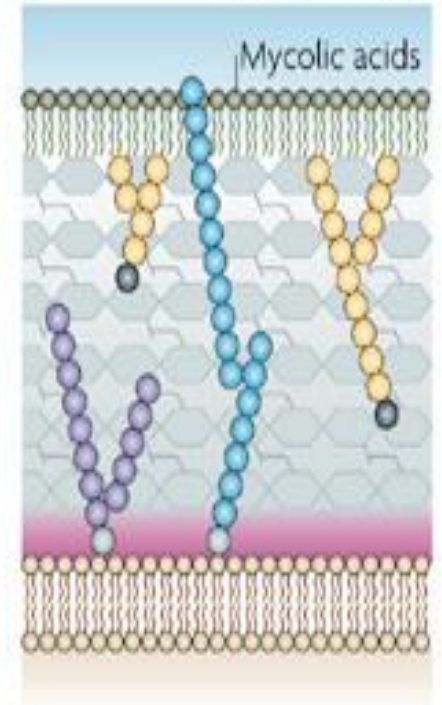
Streptococcus pneumoniae



Bacillus anthracis



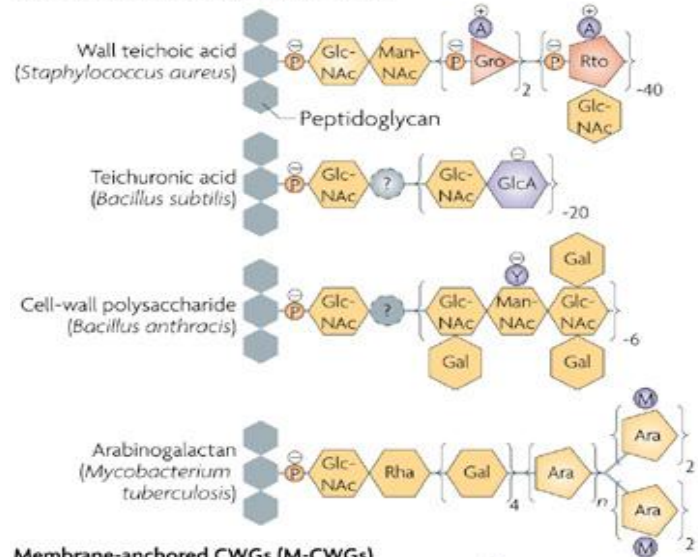
Mycobacterium tuberculosis



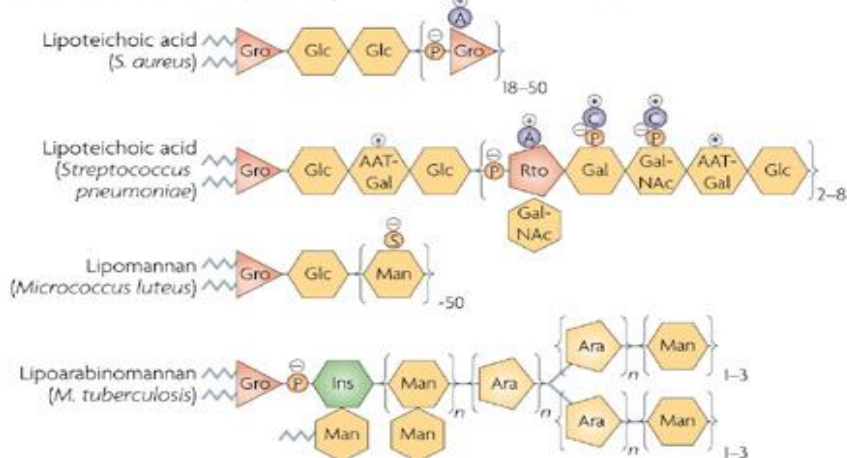
Nature Reviews | Microbiology

Schematic structures of selected CWGs.

Peptidoglycan-anchored CWGs (P-CWGs)



Membrane-anchored CWGs (M-CWGs)



- Non-glycosyl residues: A, D-alanine; C, choline; M, mycolic acid; P, phosphate; S, succinate; Y, pyruvate. Glycosyl residues: AAT-Gal, 2-acetamido-4-amino-2,4,6-trideoxy-D-galactose; Ara, arabinose; Gal, galactose; GalNac, N-acetylgalactosamine; Glc, glucose; GlcA, glucuronic acid; Gro, glycerol; Ins, inositol; Man, mannose; ManNac, N-acetylmannosamine; Rha, rhamnose; Rto, ribitol.

S. pneumoniae-Virulence Factors and Disease

- *S. pneumoniae* (C-substance)
 - most common case of bacterial pneumonia
 - Not usually the cause of a primary infection
 - Alcoholism, malnutrition, viral infections, anesthesia
 - Sinusitis, otitis media, bacteremia
 - most common cause of infectious disease deaths in the USA
 - most common cause of bacterial meningitis
 - 40% mortality

S. pneumoniae-Treatment, Prevention and Control

- Penicillin became drug of choice.
 - <1990 only 5% of U.S. strains considered resistant.
- Now resistant to multiple antibiotics.
 - Today 33% of U.S. resistant to penicillin.
- Control based on anti-capsular vaccine development.
 - 7-valent conjugated pneumococcal vaccine is recommended for children <2 years of age.
 - 23-valent recommended for at-risk adults.
 - 94% of all strains isolated from infected patients either are included in the vaccine or are serologically related to the 23-valent serotypes.

6'6"

five criminals . one line up . no coincidence

6'0"

5'6"

5'0"

4'6"

4'0"

3'6"

3'0"



The Usual Suspects

The Usual Suspects

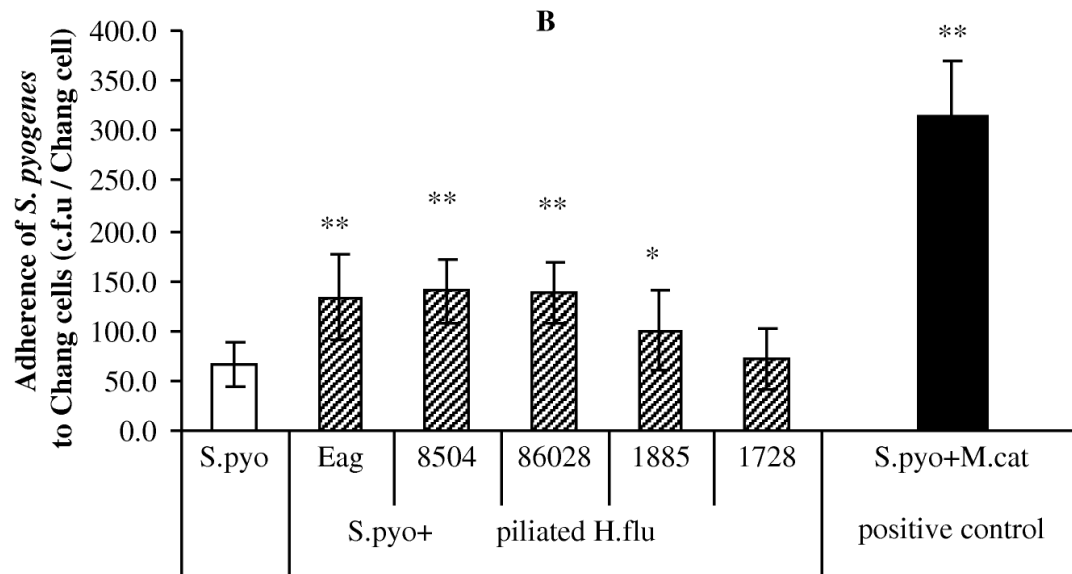
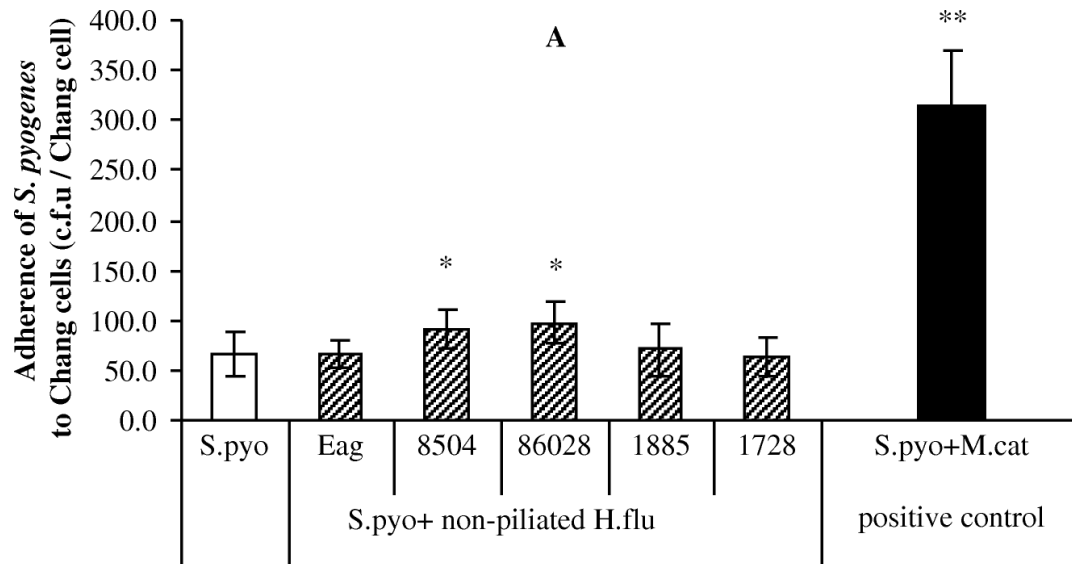
- 'The interaction between bacteria and their ecological niche can impact colonization.'

- Xu et al.
Scandinavian J.
Infect. Dis. 2009.
1:1-8

- *S. pyogenes*
 - *S. pneumoniae*
 - *Moraxella catarrhalis*
 - *Haemophilus influenzae*
- Most common bacterial species in the upper respiratory tract.

Xu et al. Scandinavian
J. Infect. Dis.
2009. 1:1-8

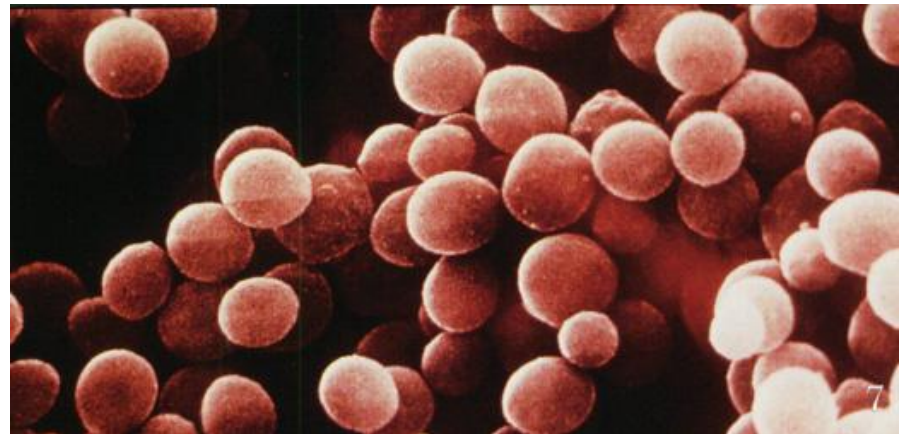
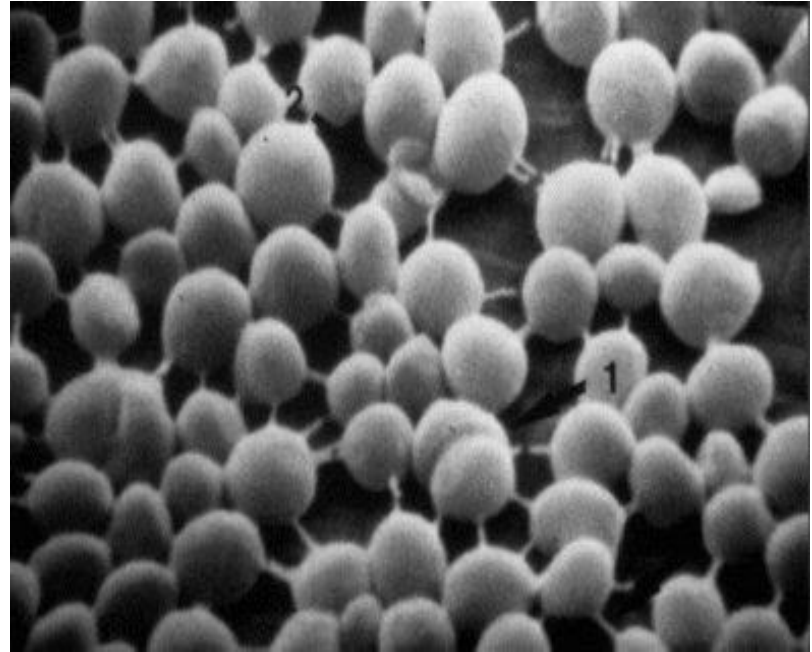
Effect of *H. influenzae* on adherence of *S. pyogenes* to human Chang cells .

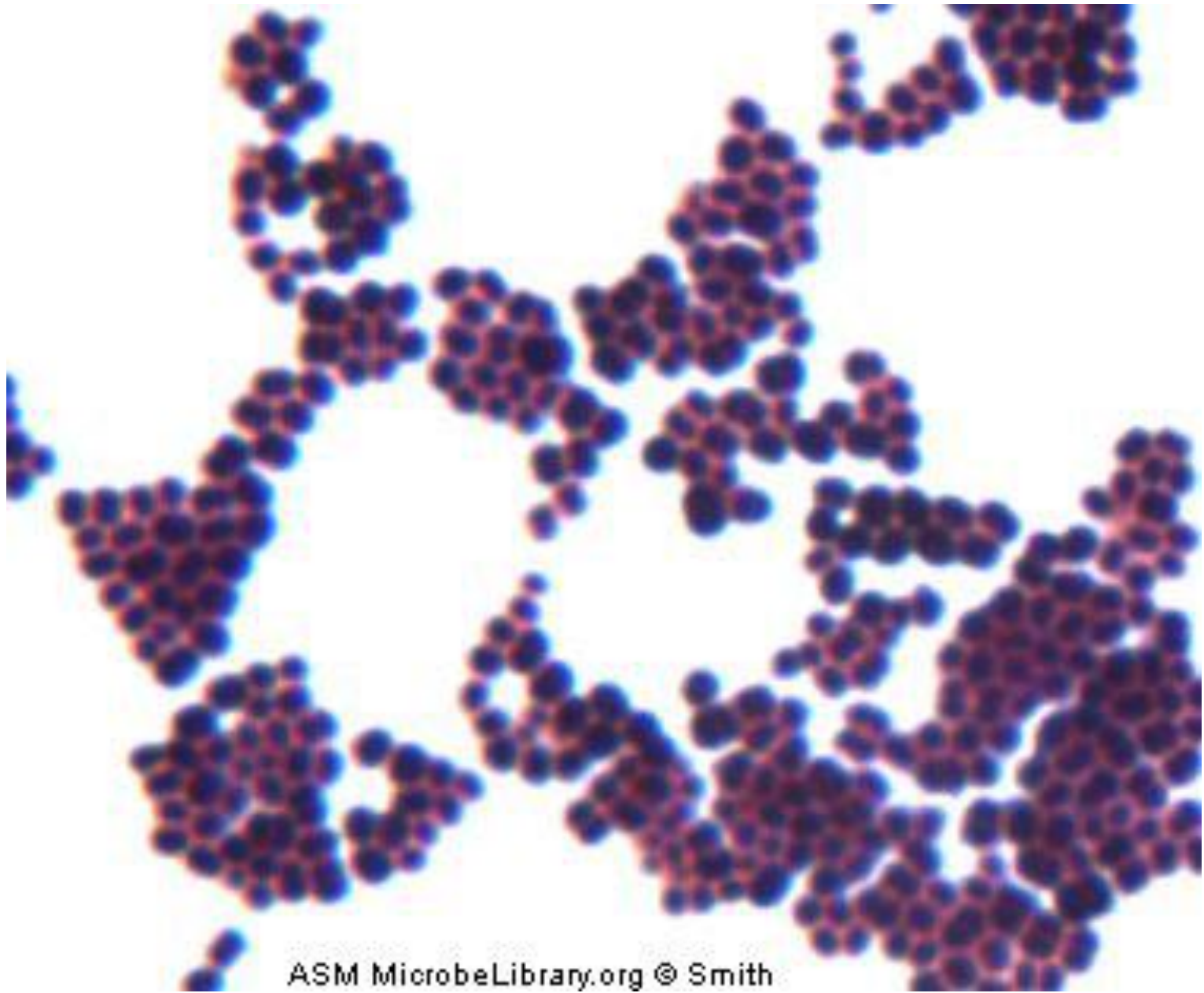


Staphylococcus aureus

*"Thou art a boil,
A plague sore, an
embossed carbuncle
In my corrupted
blood."*

**William Shakespeare
(1564-1616), *King
Lear***





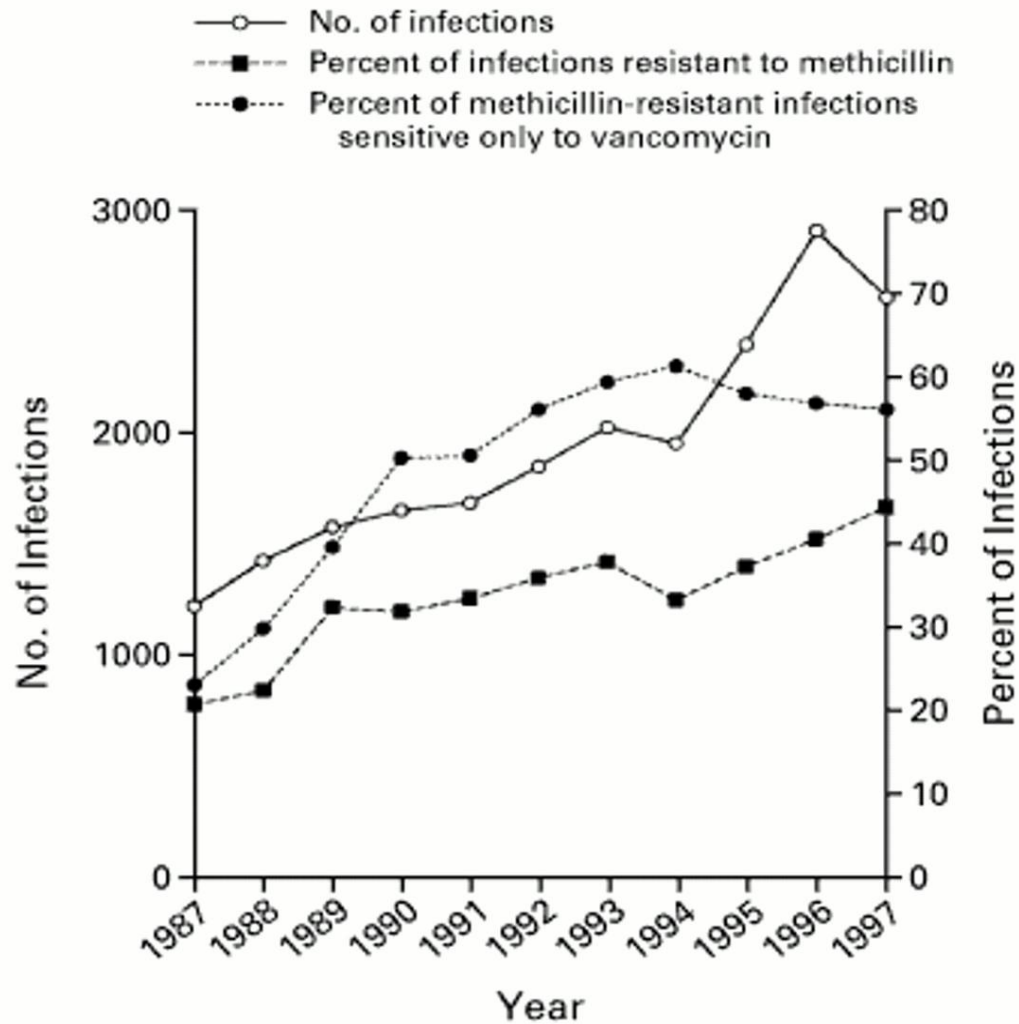
ASM MicrobeLibrary.org © Smith

The Reemergence of
Staphylococcus aureus:
infection, virulence and
immune evasion

Rise in *S. aureus* Infections

- 8987 MRSA reported between July 2004-December 2005.
 - 5250 (58.4%); community onset
 - 2389 (26.6%); hospital acquired
 - 1234 (13.7%); community-associated
 - 114 (1.3%); unclassified
- 2005: 94,360 invasive cases of MRSA; 18,650 resulted in death.
 - Incidence rate of 31.8/100,000 compared to 1/100,000 for *S. pneumoniae* and *H. influenzae*.

Temporal Changes in *S. aureus* Disease



Use of Antibiotics in Human Medicine and Resistant Gene Transfer

- In June 2002 a milestone was passed...
 - **Vancomycin** was the last line of defense against *Staphylococcus sp.* and *Streptococcus sp.*
- The vancomycin barrier began to crack when *Enterococcus sp.* began to become vancomycin resistant.
- In 2003, the first fully-resistant strain of *S. aureus* was reported.
 - this strain was isolated from a dialysis patient
 - such patients often develop mixed infections
 - in this case, both *S. aureus* and *E. faecium* were isolated from the patients blood.

Staphylococcus aureus

- Can be cultured from dry surfaces after several months.
- Relatively heat resistant.
- Can survive high salt concentrations

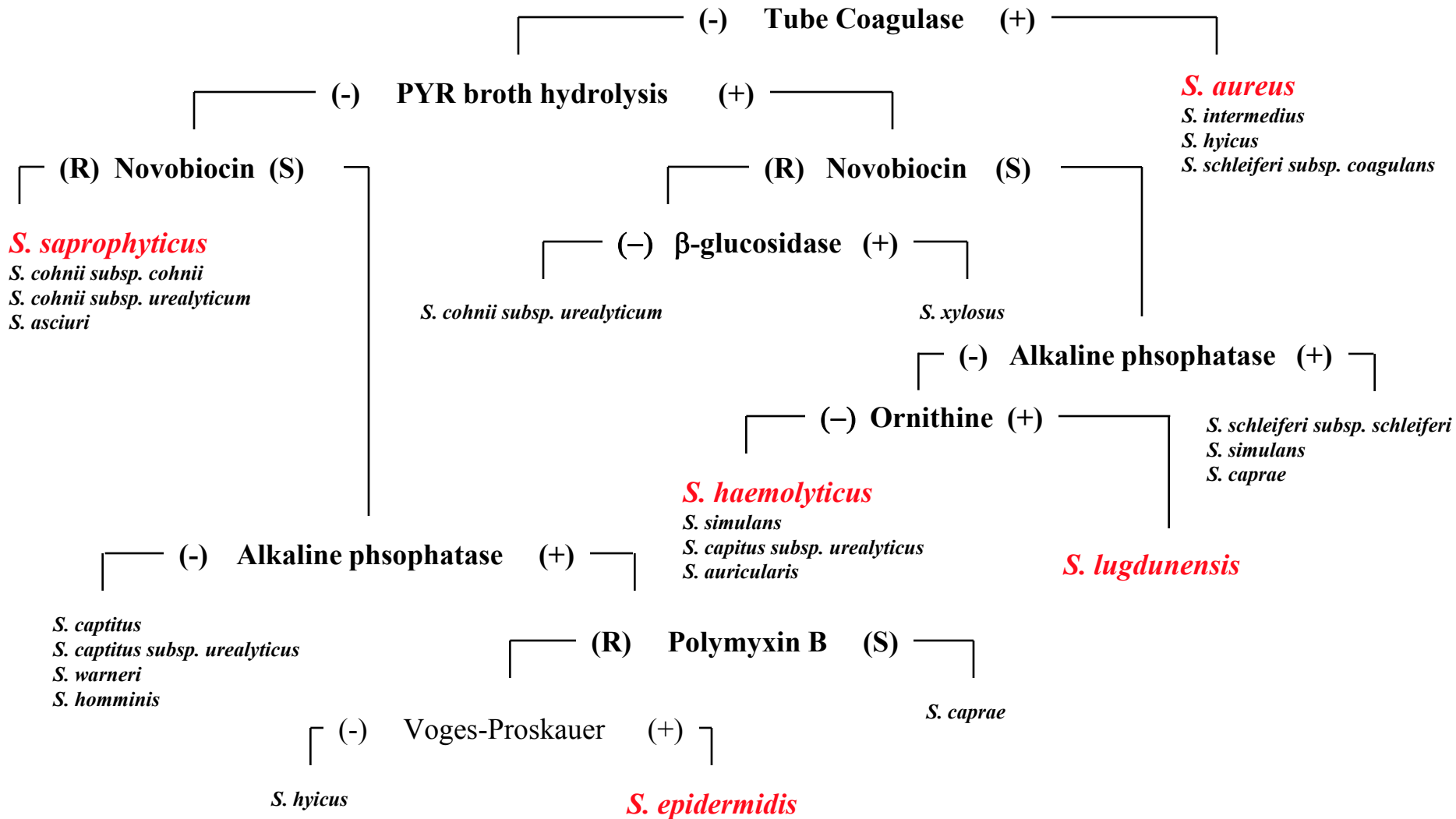


Staphylococcus aureus

- Gram + cocci, **benign** (?!?), symbiotic, and commonly found in the normal flora of the skin.
- Distinguished from other staphylococci on the basis of the gold pigmentation, positive **coagulase** tests, mannitol fermentation, **catalase**, and deoxyribonuclease production.



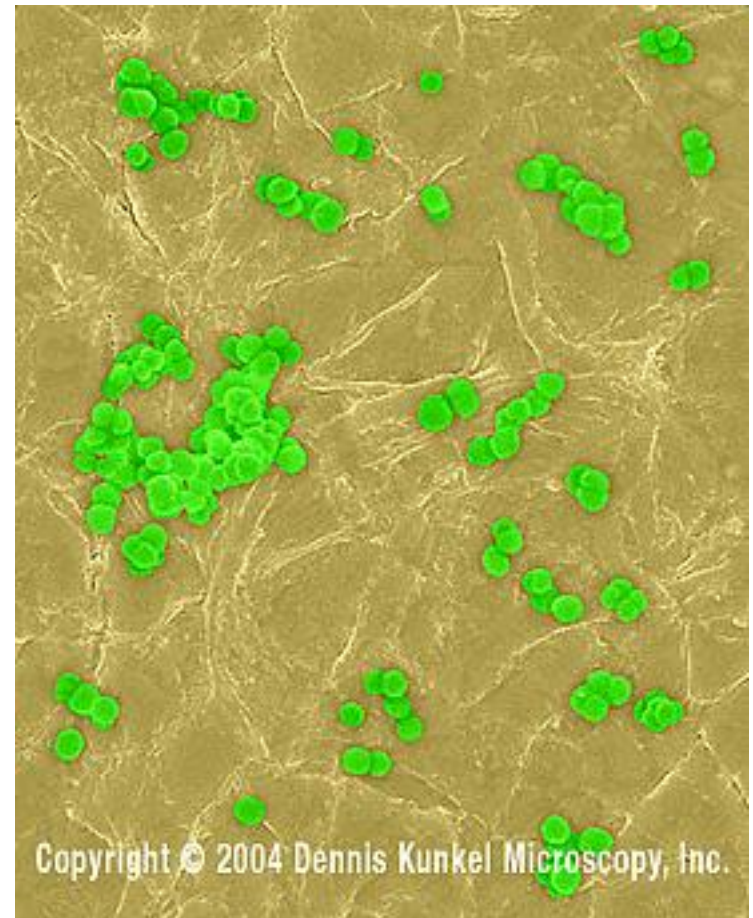
Staphylococcal Species



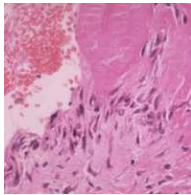
What Makes *S. aureus* a Formidable Pathogen?

- *S. aureus* is the agent of numerous types of infections, syndromes, and toxinosis
- *S. aureus* produces numerous types of virulence factors that are coordinately regulated by adaptive systems
- *S. aureus* has the ability to persist in host cells
- Pan-antibiotic resistant strain of *S. aureus* have emerged

Anytime anywhere...



S. aureus and Disease



Endocarditis



Cellulitis



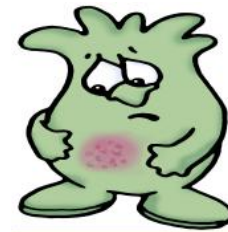
Endophthalmitis



Scalded Skin Syndrome



Toxic Shock Syndrome



Food Poisoning



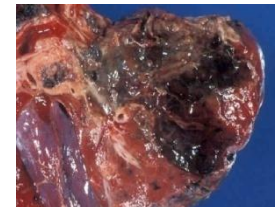
Osteomyelitis



Boils



Impetigo



Pneumonia

What Makes *S. aureus* a Formidable Pathogen?

- *S. aureus* is the agent of numerous types of infections, syndromes, and toxinosis.
- *S. aureus* produces numerous types of virulence factors and immunomodulatory molecules that are coordinately regulated by adaptive systems.
- *S. aureus* has the ability to persist in host cells.
- Pan-antibiotic resistant strain of *S. aureus* have emerged.

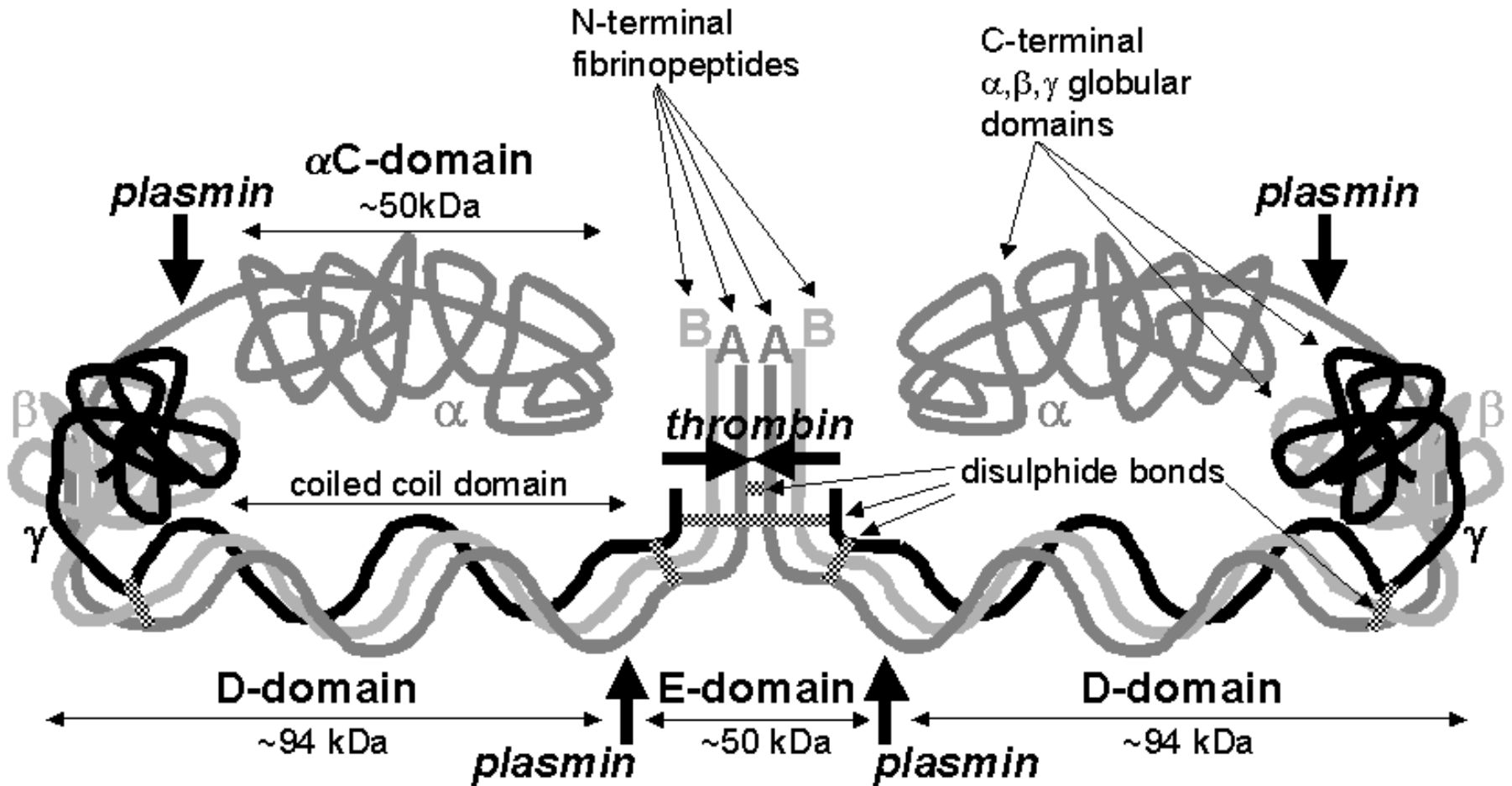
Staphylococcus aureus

- **Catalase:** All *S. aureus* produce hydrogen peroxide that is converted to oxygen and water by catalase.
- Suggested that catalase contributes to the pathogenicity of staphylococci by splitting toxic **oxygen radicals** important in killing phagocytosed bacteria.

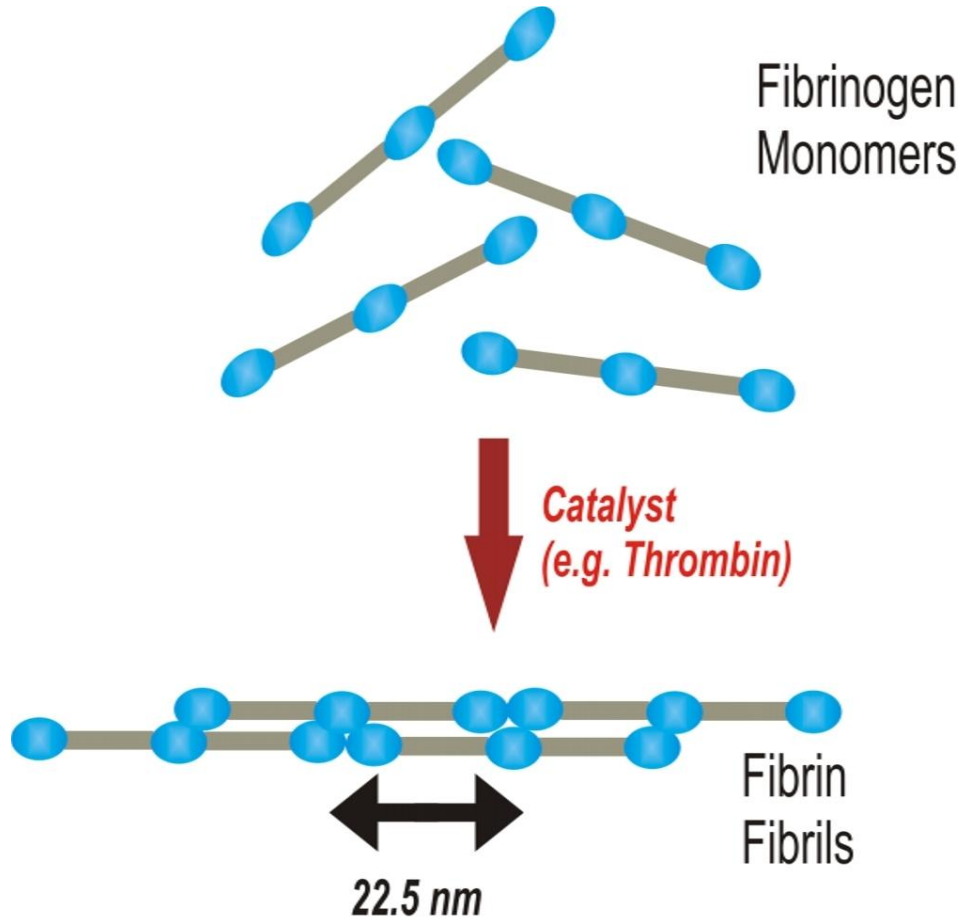
Staphylococcus aureus

- *Coagulase*: Both cell-bound and soluble coagulase mediated coagulation by steps that differ from those that participate in normal thrombosis...
- Virulence...?

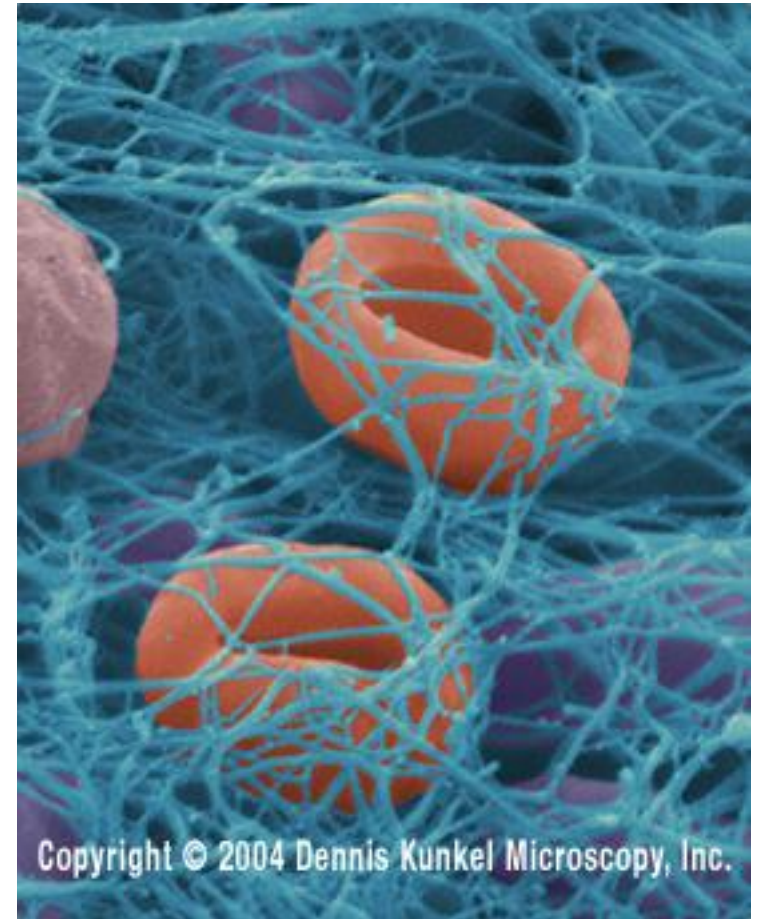
Coagulation



Coagulation



Coagulation



Virulence Factors & Virulence Factor Regulation

Staphylococcal Virulence Factors

TOXINS

- α -toxin
- β -toxin
- δ -toxin
- Enterotoxins A-J
- Exfoliative toxin A and B
- Panton-Valentine toxin/
 γ -toxin
- Toxic Shock Syndrome
Toxin

ENZYMES

- β -lactamase
- Catalase
- Coagulase
- Cystein protease
- V8 protease
- Metalloprotease/aureolysin
- Staphopain

- Lipase
- Nuclease
- Thermonuclease
- PI-phospholipase C
- Staphylokinase
- Hyaluronidase
- Lipase/esterase
- Serine-like proteins

SURFACE PROTEINS (MSCRAMMS)

- Clumping factor A/B
- Collagen binding protein
- Fibrinogen-binding protein
- Fibronectin-binding protein
- Lactoferrin-binding protein
- Laminin binding protein
- Extracellular matrix-binding protein
- Vitronectin-binding protein
- Elastin-binding protein
- Bone sialoprotein-binding protein
- Serine-aspartate repeat protein

MIMS

- Efb, Ecb, SCIN
- Map
- Protein A
- CHIPS/FLIPR
- SAK

Types of Toxins

- I- Membrane-active agents
- II-Pyrogenic toxin
superantigens (PTSAgs)
- III-Exfoliative toxins (ETs)

Staphylococcal Virulence Factors

TOXINS

- α -toxin
- β -toxin
- δ -toxin
- Enterotoxins A-J
- Exfoliative toxin A and B
- Panton-Valentine toxin/
 γ -toxin
- Toxic Shock Syndrome
Toxin

- Lipase
- Nuclease
- Thermonuclease
- PI-phospholipase C
- Staphylokinase
- Hyaluronidase
- Lipase/esterase
- Serine-like proteins

ENZYMES

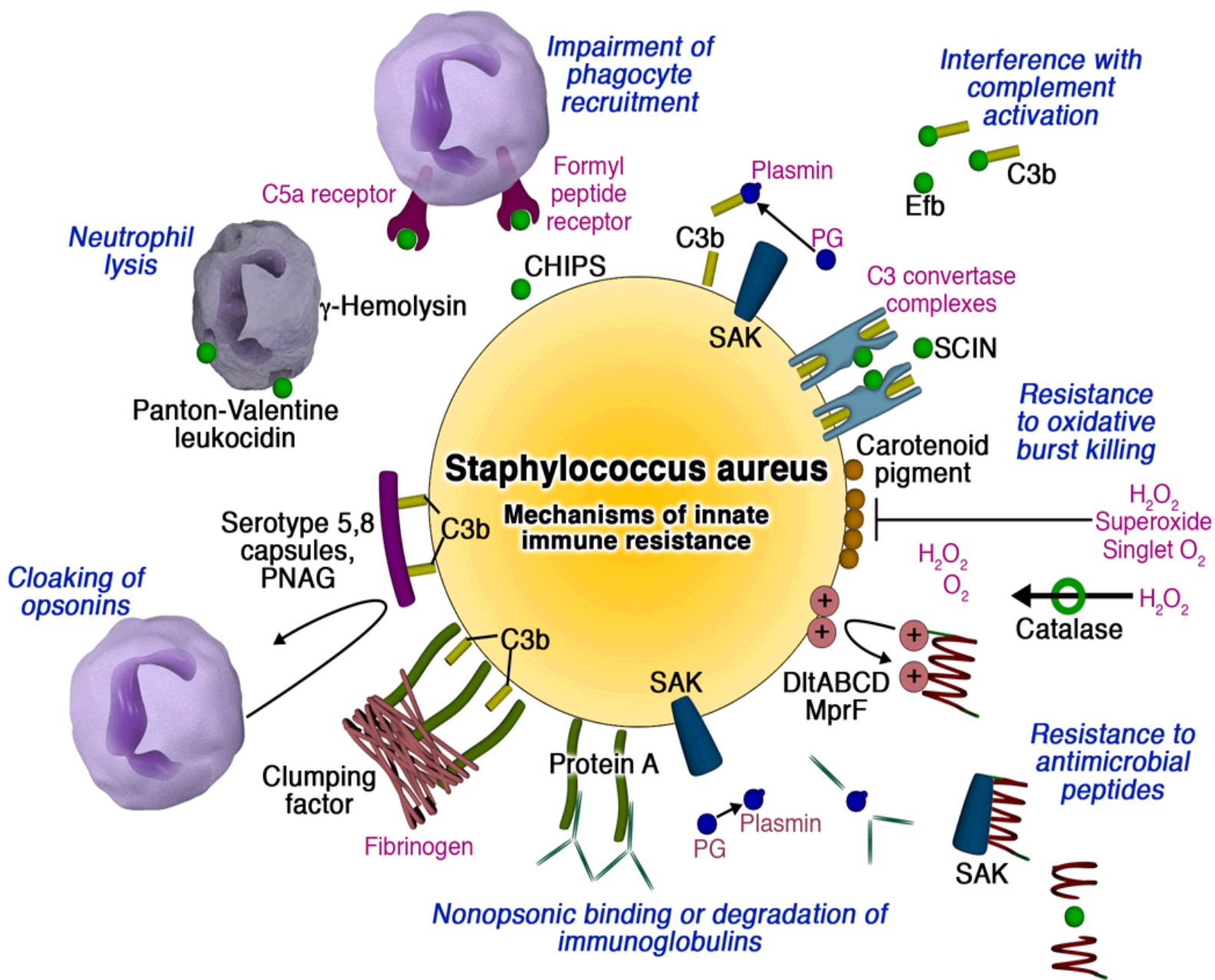
- β -lactamase
- Catalase
- Coagulase
- Cystein protease
- V8 protease
- Metalloprotease/
aureolysin
- Staphopain

SURFACE PROTEINS (Adhesins)

- Clumping factor A/B
- Collagen binding protein
- Fibrinogen-binding protein
- Fibronectin-binding pr
- Lactoferrin-binding protein
- Laminin binding protein
- Extracellular matrix-binding protein
- Vitronectin-binding protein
- Elastin-binding protein
- Bone sialoprotein-binding protein
- Serine-aspartate repeat protein

MIMS

- Efb, Ecb, SCIN
- Map
- Protein A
- CHIPS/FLIPr
- SAK

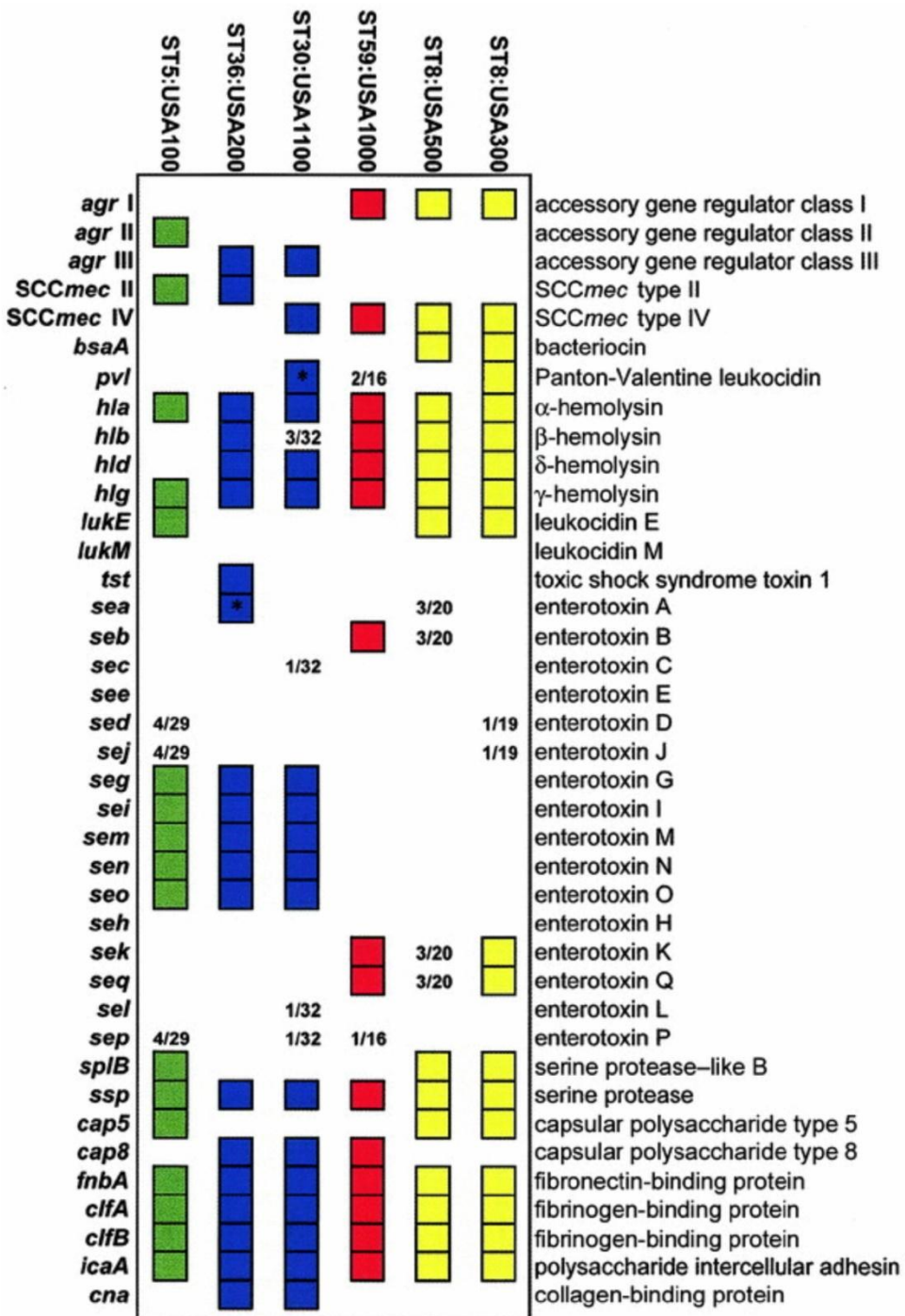


Adapted from Victor Nizet, MD

Then USA300...

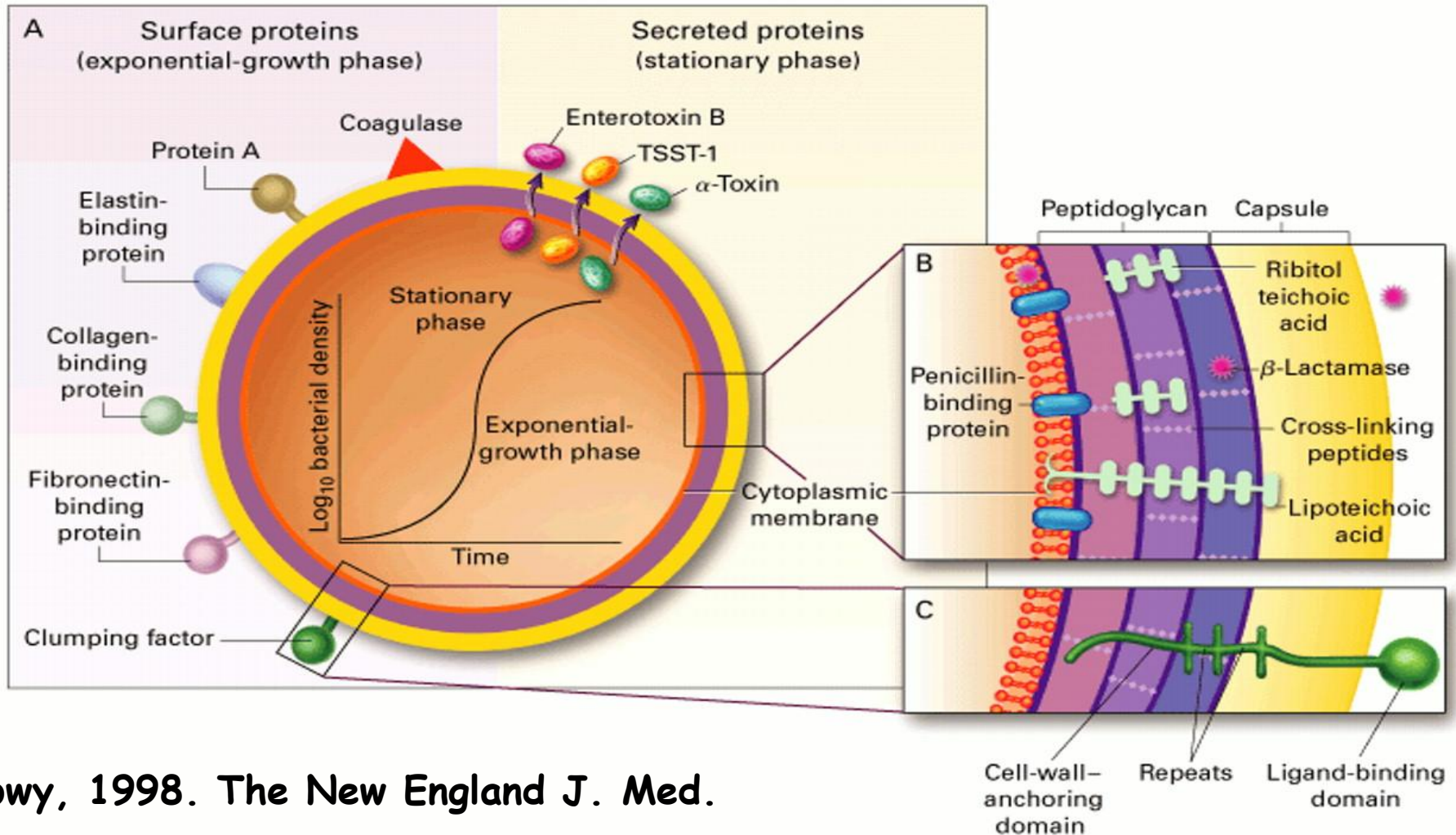
S. aureus strain USA300

- Plasmids
 - pUSA01
 - cryptic
 - pUSA02
 - *tetK*
 - tetracycline resistance
 - pUSA03
 - *ermC, ileS*
 - resistance to macrolides, lincosamides, streptogramin B., and mupirocin
- Mobile genetic elements
 - SCCmec Type IV
 - Methicillin resistance
 - ciprofloxacin
 - ACME type I
 - Arginine deaminase pathway
 - SaPI5
 - *seq2, sek2*
 - ϕ SA2usa
 - *lukS-PV, lukF-PV*
 - ϕ SA3usa
 - *sak, chp*



Diversity of Virulence Factors between *S. aureus* isolates.

Structure of *S. aureus*--A Temporal Pattern of Virulence Factor Expression *in vitro*

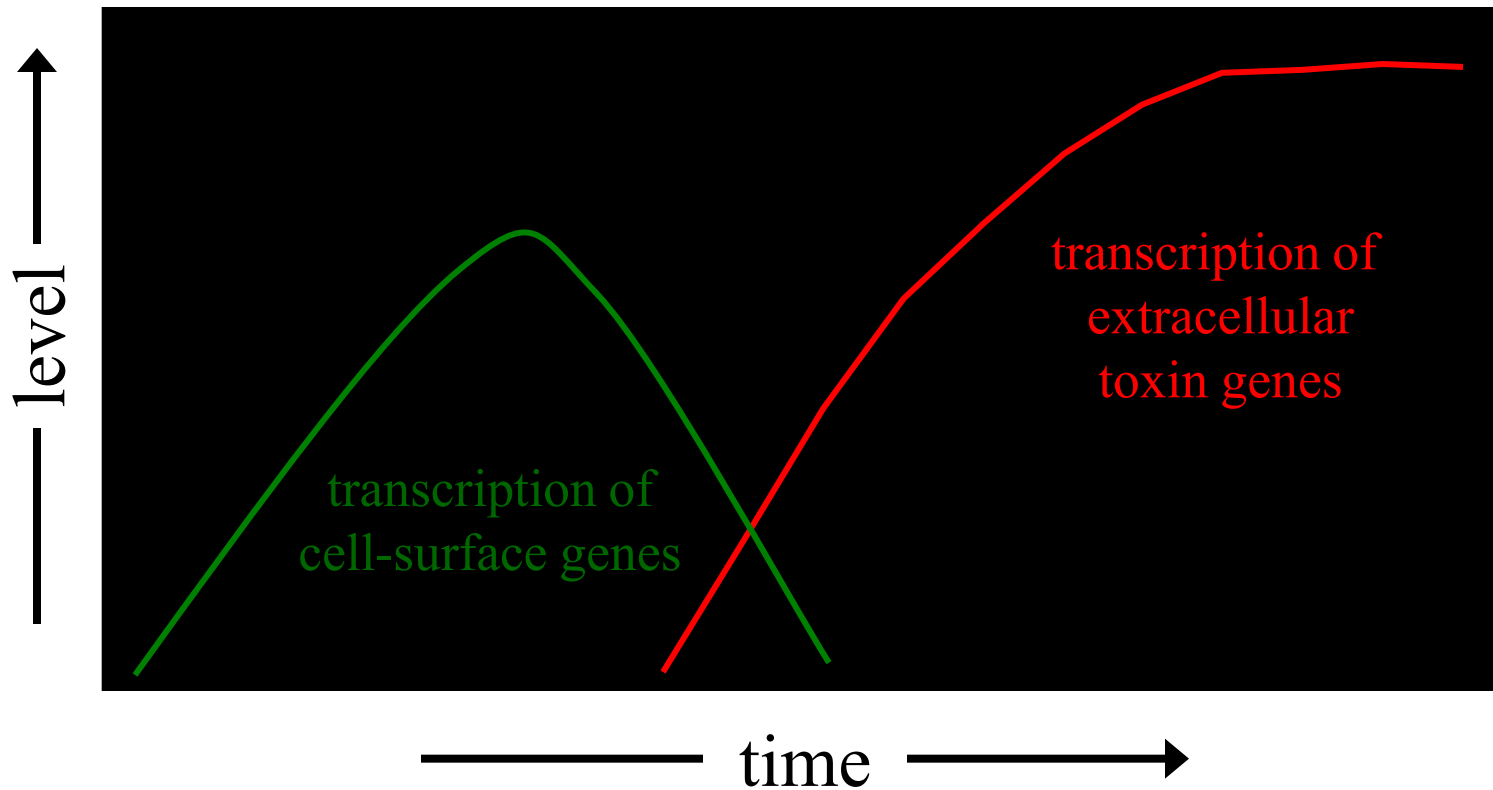


Lowy, 1998. The New England J. Med.

A Temporal Pattern of Virulence Factor Expression *in vitro*

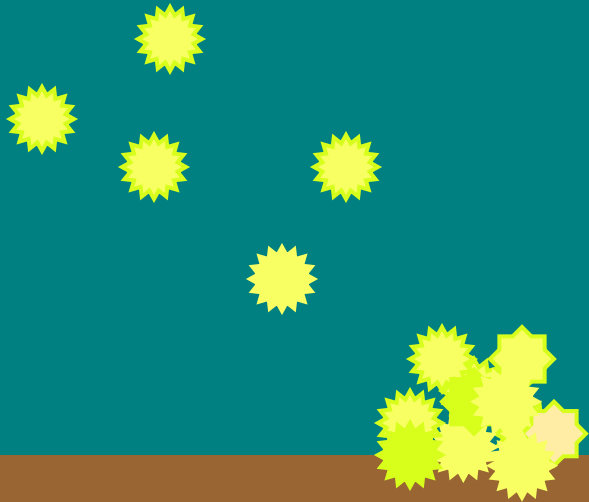
- Early studies in laboratory culture identified a characteristic pattern of cell-surface and extracellular virulence factor expression
- In the **exponential phase** of bacterial growth - the bacteria express select cell-surface virulence factors and the expression of extracellular virulence factors is down regulated
- **Post-exponential** phase of bacterial growth - the production of cell-surface virulence factors ceases and the genes encoding extracellular virulence factors are de-repressed
- Spontaneous mutations that affected the production of multiple virulence factors suggested regulation by a common mechanism

Temporal Expression of Virulence Factors *in vitro*

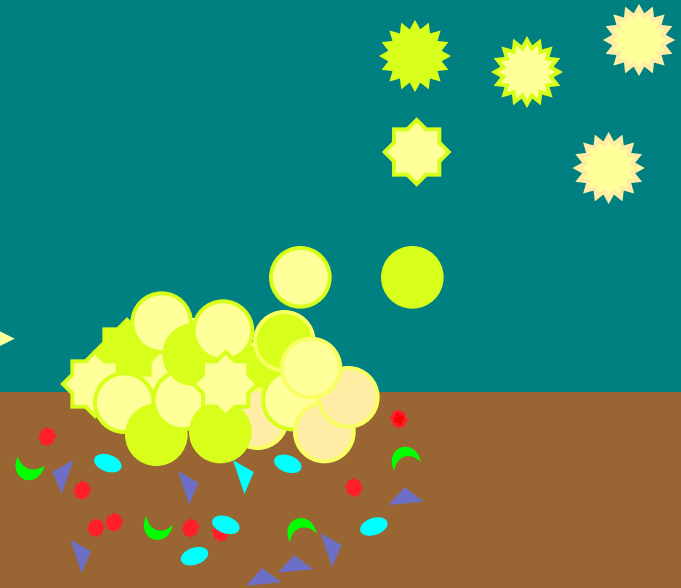


Toxin Regulation *in vivo*?

low cell density



high cell density

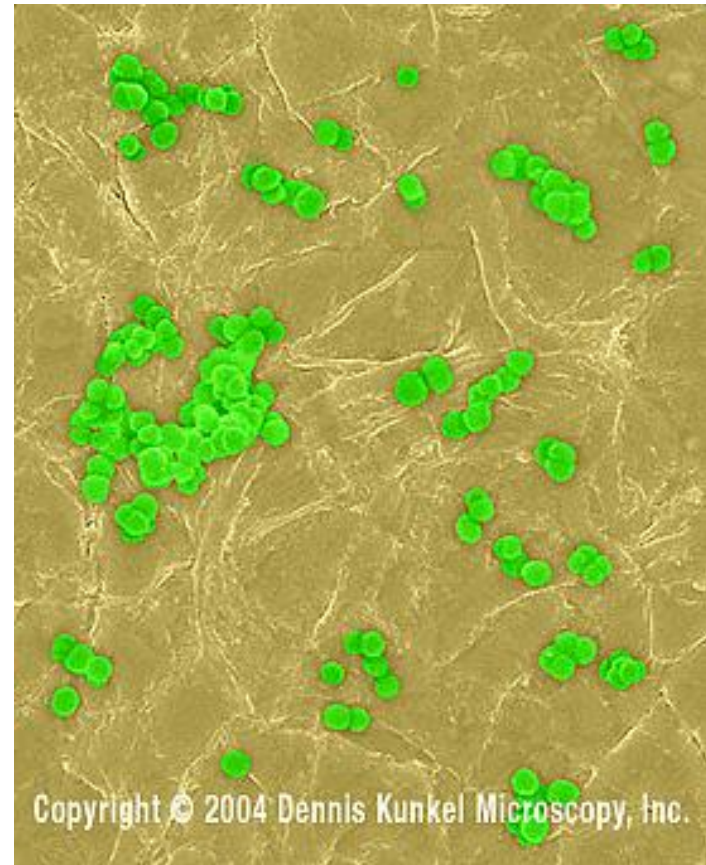


Surface proteins expressed
Toxins and enzymes down-regulated

Surface proteins down-regulated
Toxins and enzymes expressed

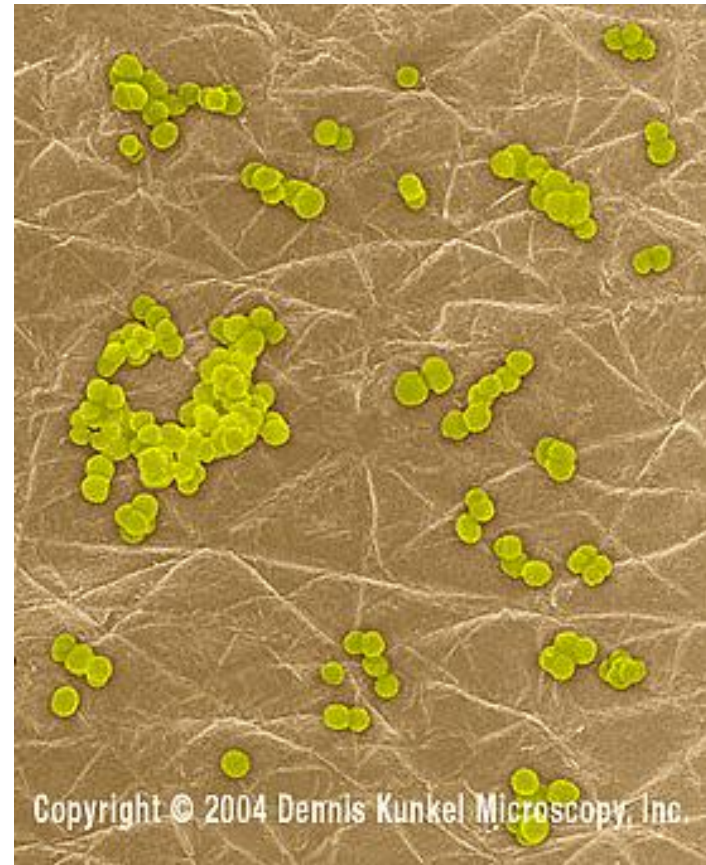
Epidemiology of *S. aureus*

- Shortly after birth, staphylococci from the contiguous human environment colonize the infant's umbilical stump, skin, perineum, and sometimes the GI tract.

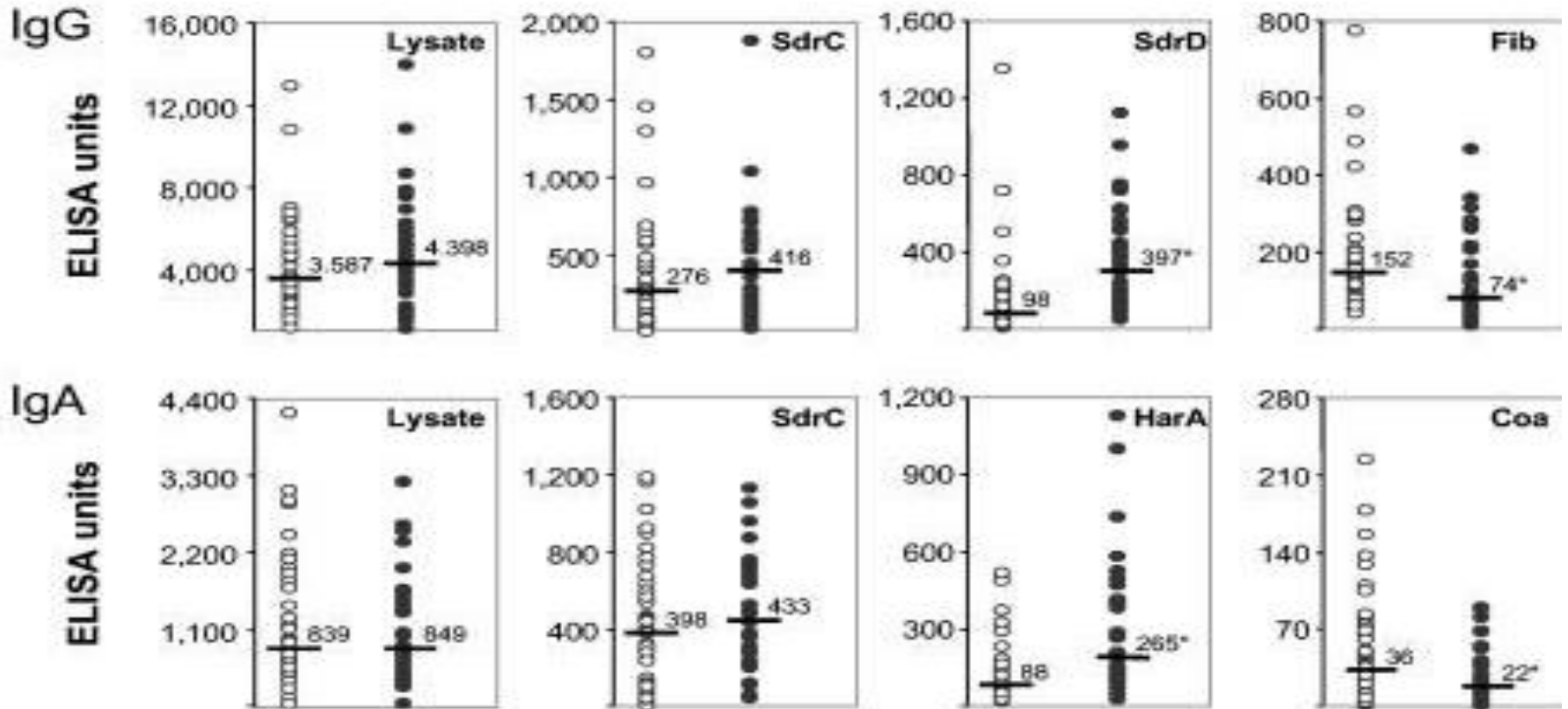


Epidemiology of *S. aureus*

- **Humans are a natural reservoir.**
- 30-50% of healthy adults are colonized.
- 10-20% persistently colonized.
 - nasal carriage
- Both MSSA and MRSA are colonizers.
- Colonized individuals are at greater risk for subsequent infections.



IgG and IgA antibody levels in sera from **healthy and infected** individuals were determined by ELISA with recombinant proteins.

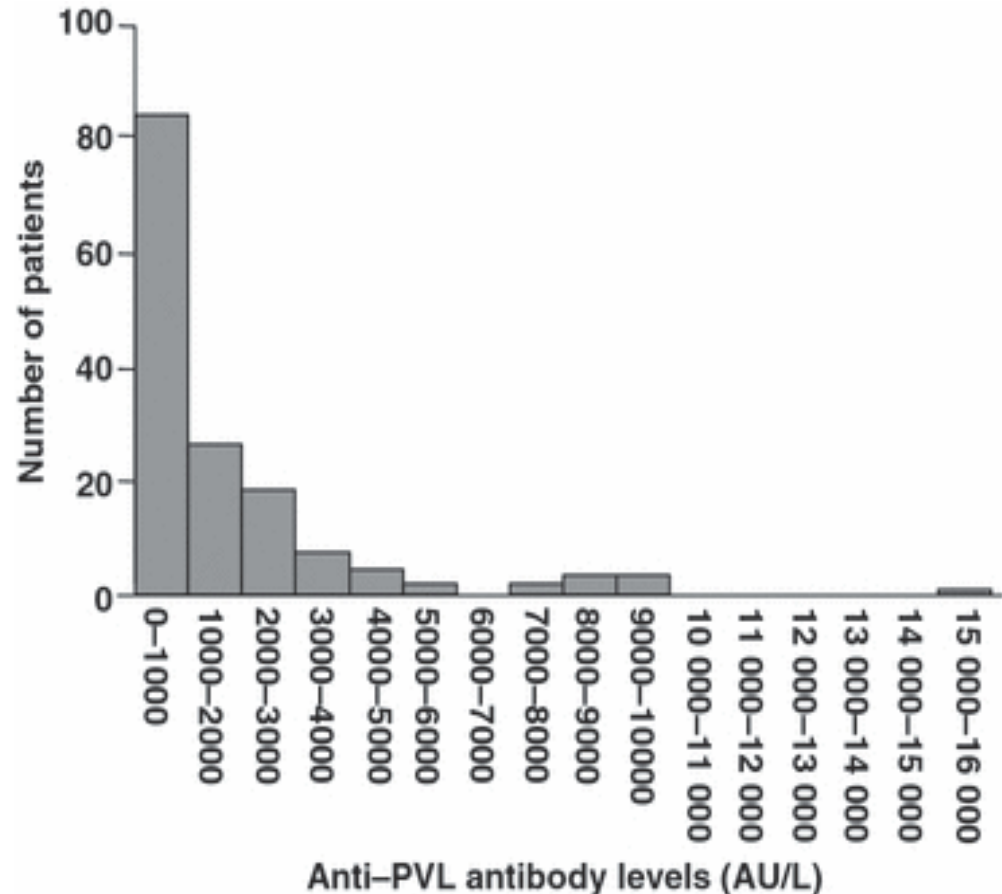


Dryla et al. Clin Diagn Lab Immunol. 2005 March;
12(3): 387-398.

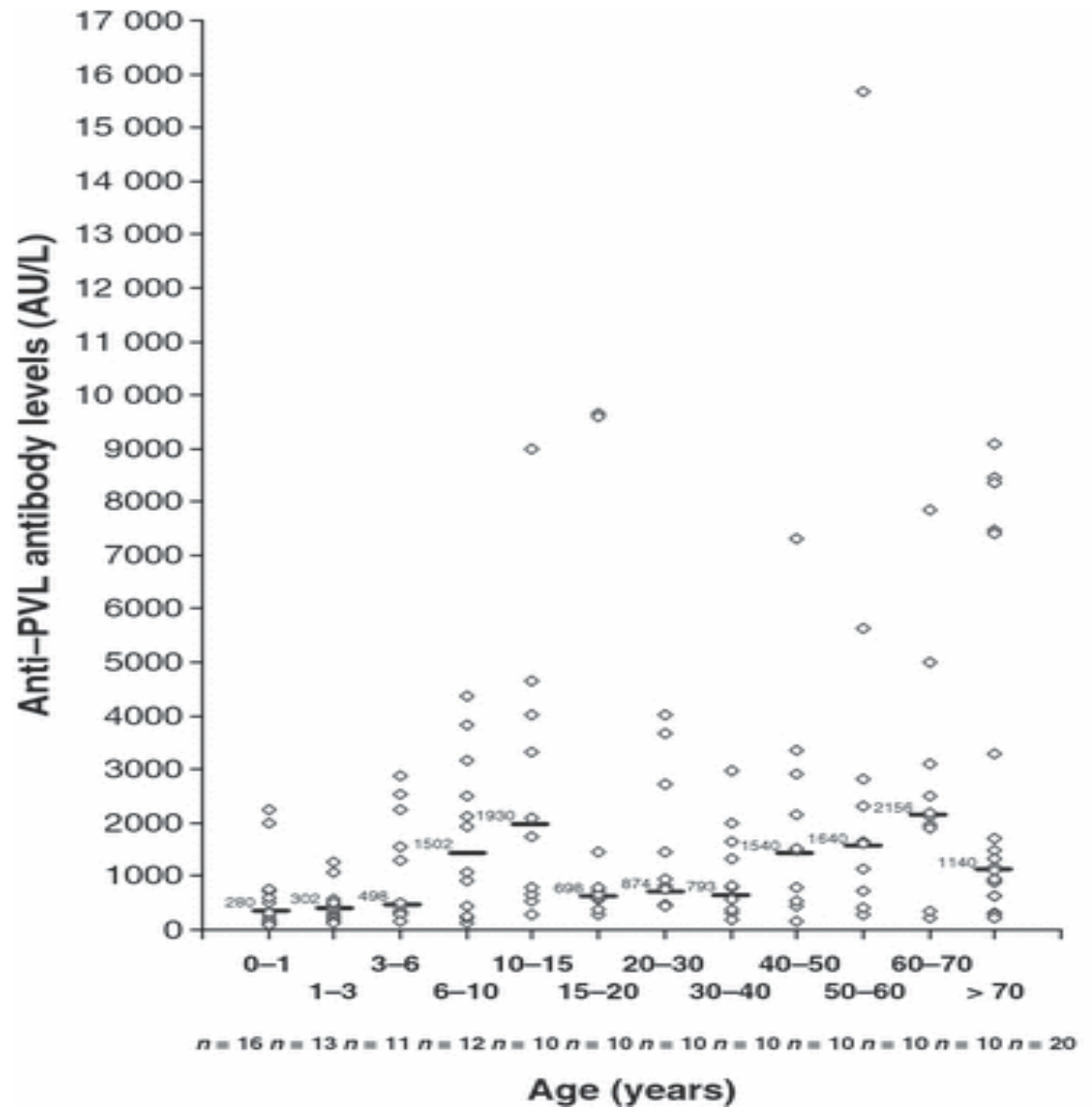
doi: 10.1128/CDLI.12.3.387-398.2005.

Distribution of anti-Panton-Valentine leukocidin (PVL) antibody levels in patients with **no evidence** of *Staphylococcus aureus* infection.

- The results were expressed in arbitrary units per liter (AU/L); 1000 arbitrary units correspond to the amount of anti-PVL antibodies contained in a solution of commercial human polyclonal immunoglobulins at 12.5 g/L, which is the physiological human immunoglobulin serum concentration.



Distribution of anti-Panton-Valentine leukocidin (PVL) antibody levels in the different age groups of patients with no evidence of *Staphylococcus aureus* infection. Bars show median values.



M. Croze et al. 2009. Clin. Microbiol. Infec.
<http://www3.interscience.wiley.com/journal/121636951/abstract>

Epidemiology of *S. aureus*

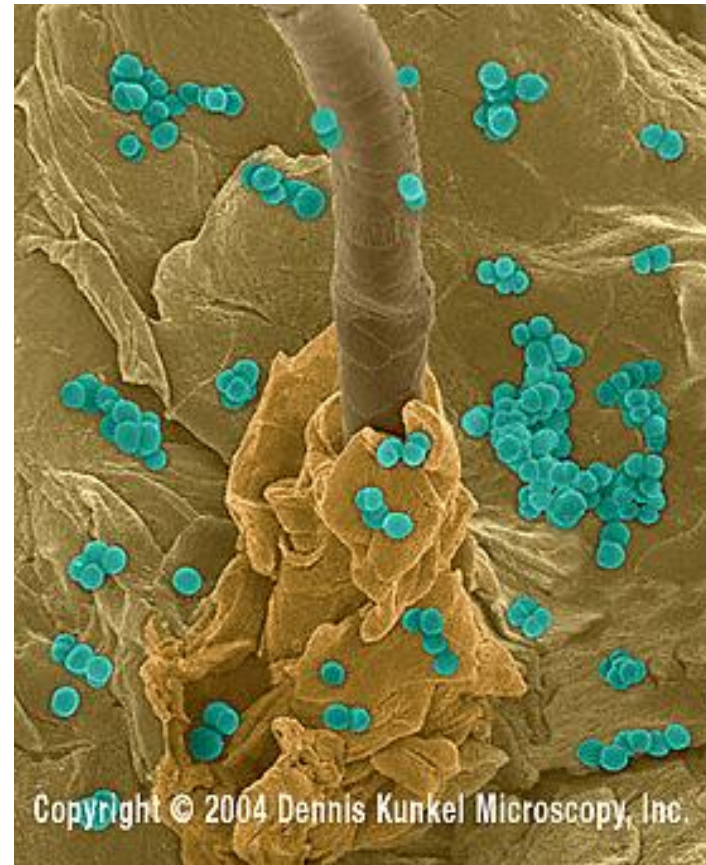
- Other risk factors:
 - i.v. drug users
 - **Diabetes**
 - hemodialysis patients
 - surgical patients
 - immunodeficient patients
 - patients with deficits in leukocyte function
 - poor hygiene
 - **insect bites**
 - cutaneous disorders (eczema and acne)

Transmission

- Colonized individuals are at greater risk (autoinfection).
- **Nosocomial** infections are acquired through exposure to the hands of health care workers after they have been transiently colonized with staphylococci from their own reservoir or from contact with an infected patient.
- Outbreaks may result from exposure to a single long-term carrier or environmental sources.
- **CA-MRSA and CA-MSSA...later.**

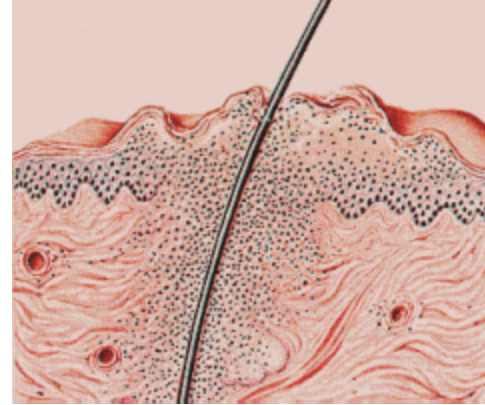
Localized Infections

- **MOST INFECTIONS CENTERED AROUND HAIR FOLICLES.**
- **Systemic features are usually absent...**



Folliculitis

- Acute inflammation
- Pink to red papules
- Rapidly extrude pus from the orifice of the involved follicle.
- Resolution may be spontaneous or by local antiseptic measures.
- If the infections spreads...



From folliculitis to a boil

- ...to perifollicular subcutaneous tissue, a furuncle (boil) may result.
- Raised lesions up to 1 cm in diameter.
- If not drained by incision, furuncles break down centrally and discharge pus.



From a boil to a carbuncle

- Results from subsequent spread of infection more deeply along facial planes.
- Very dangerous.
- May affect deeper structures.
- Surgical intervention necessary.
- Also referred to as **cellulitis**.



Massive periorbital cellulitis caused by *S. aureus* with thrombosis of the cavernous sinus.



Impetigo

- Epidermis infection
- Similar to a boil.
- Very contagious.
- **Primarily in children.**
- Lesions begin as minute red macules that rapidly become vesicular and contain cloudy fluid or pus on an erythematous base.
 - 80% *S. aureus*
 - 10% streptococcal
 - 10% mixed
- Antiseptic measures important in prevention/containment



Differential diagnosis early includes herpes and varicella.

Staphylococcal Scalded Skin Syndrome (SSSS)

- Ritter's disease
- Most typical in newborns (and less common in older children and adults--**lymphoma**).
- Disease begins with perioral edema.



Staphylococcal Scalded Skin Syndrome (SSSS)

- Flaccid bullae rupture releasing a clear, sterile fluid.
- Clinically uninvolved skin can be made to exfoliate by friction.
- The disease is a toxic response of the epidermis to exfoliatin.
- Mortality in infants is 3-5%.



Staphylococcal Scalded Skin Syndrome (SSSS)

- Exfoliatins are soluble exotoxins made by some *S. aureus* strains which bind to **filaggrins**, a group of proteins associated with keratohyaline granules in the skin.
- The immaturity of the **intraepidermal cellular attachment** mechanism are thought to explain the preponderance of SSSS in newborns (toxin less rapidly excreted in the urine).

Toxic Shock Syndrome

"In general the adaptive relationship between microorganism and host is effective only for the precise circumstances under which adaptation evolved--in circumstances which constitute physiological normalcy for the host concerned. Any departure from this normal state is liable to upset the equilibrium and to bring about a state of disease."

--René Dubos, 1961

During the summer of 1982, an angry Dr. Michael Lange took the podium at the GRID conference in New York city and decried the paucity of concern and research funds from the U.S. government. With 177 GRID deaths recorded, he said, the mystery was receiving virtually no attention. In contrast, TSS, which, he claimed, had killed only 85 women in four years, was grabbing headlines, national attention, and federal dollars...

The 'Mystery' Behind TSS...

- 1936-Earle Hass designs the first tampon.
- Despite a widespread hue and cry about the immorality of such a device, Haas had no trouble selling his patent to Tampax Inc.
 - It was not until the **1970s** that the National Association of Broadcasters lifted their long-standing ban on radio and television advertising of tampons, and newsmagazines followed suit.
- In the United States no federal or state health agencies oversaw tampon production.
- By the 1960s, tampons were in widespread use wherever in the world women could afford them.

The Mystery Behind TSS...

- Competition escalated radically.
- Absorbency
 - Procter & Gamble released a tampons made of polyester and plastic.
 - Dozens of different stabilizing fibers were used, including asbestos.
- By 1975, the five tampon manufacturers were spending \$billions on advertising.
- Among the synthetic materials used in marketed super absorbent tampons in 1978-81 were polyurethane, polyester, collagen, polyvinly alcohol, acetyl cellulose, and **carboxymethyl cellulose (CMC)**.
- In 1979, P&G released a tampon comprised of highly compressed beads of alternating polyester and CMC.
 - Rely Tampon

The Impact of Rely

- Following a massive advertising kickoff, Rely hit the North American market.
- Other manufacturers retaliated immediately, marketing Assure, MaxiSorb, etc.
- Absorb **20x their weight in fluids**.
 - holding more fluid than most women actually had in their vaginas at a given time.



"Any departure from this normal state is liable to upset the equilibrium and to bring about a state of disease."

Warnings were there...

- Tests on rabbits showed that sterilized tampons made from collagen, polyurethane, polyvinyl alcohol, or acetyl cellulose produced lesions and ulcerations in the vagina's epithelial tissue.
- Re-growth of the epithelium plummeted (20-80%).
- CMC acted as a toxin filter, **especially for *S. aureus* toxins.**
- From the moment superabsorbent tampons hit the market, there were published accounts of vaginal ulcerations, lesions, and lacerations.

Uncovering the Mystery

- 1977-pediatrician James Todd described an acute life-threatening ailment he called Toxic Shock Syndrome.
 - Children were infected between 1975-1977 with *S. aureus*.
 - One of his patients was 15-year-old girl who was at death's door for 8 days.
 - Her vaginal discharge contained a strain of *S. aureus* that was remarkable for two features:
 - resistant to the entire penicillin class of antibiotics
 - appeared to secrete some unique toxin
 - Literature suggested a similar case in 1927 (New York) and in 1941 (Baltimore).

Uncovering the Mystery

- By early 1980, there were forty-three more confirmed cases of TSS.
- Arguments about how TSS should be defined.
- From 1975-October 1980, 408 reported cases.
 - 394 were women
 - 95% were menstruating
 - 40 died
 - 1/3 had previously had a previous milder episode
- For a very long time, the concept that a **toxin was responsible** (and not the organism directly) eluded everyone.
- Rely blamed initially and taken off the market.
- Cases continued to present over time. **Why?**
- First set of 'regulations' put forth for safety.
- By January 30, 1981, 941 cases and 73 deaths.

Enter Patrick Schlivert...

- Only researcher to suspect a toxin and do something about it.
- Was denounced by the CDC as 'weaving bizarre theories in public view, failing to publish his data, and seeking the media spotlight for personal glory.'



Patrick Schlivert

- The CDC stated that, "More work needed to be done to prove that Schlievert's candidate is the actual toxin."
- Schlivert: "I don't care what the CDC says."
 - Schlivert had already isolated the toxin and proved that it was responsible for the symptoms of TSS.
 - Toxin was a Superantigen called TSST-1

TSST-1

- Superantigens can bind independently to MHC II molecules and to T cell receptors, binding to the V domain of the TCR and to the outer faces of the MHC II molecule (outside of the peptide-binding groove).

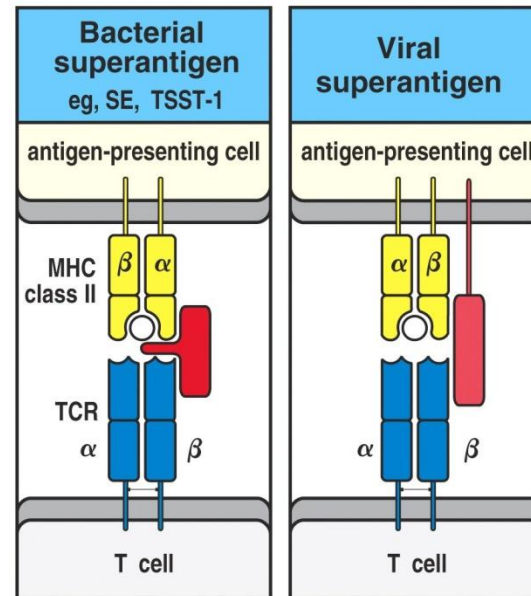


Figure 5-19 part 1 of 2 Immunobiology, 6/e. (© Garland Science 2005)

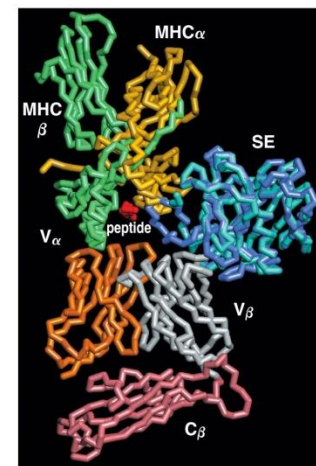
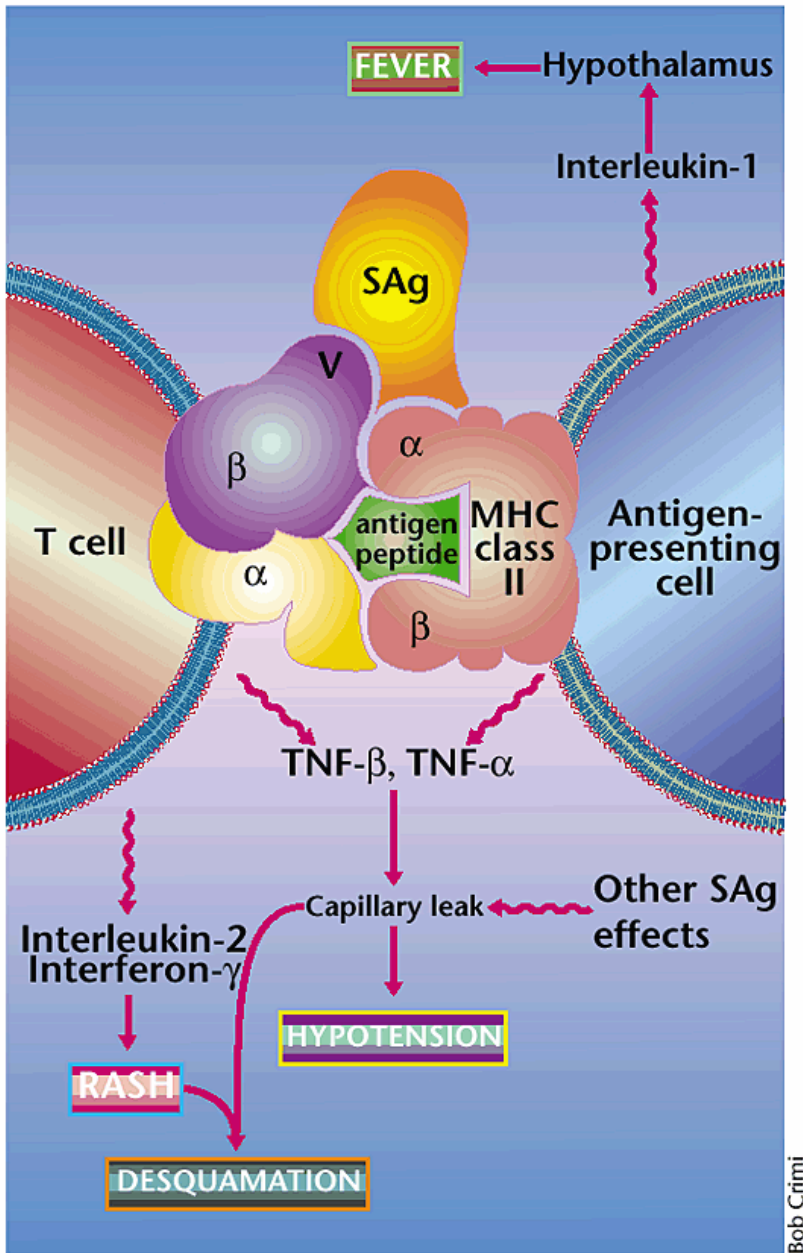


Figure 5-19 part 2 of 2 Immunobiology, 6/e. (© Garland Science 2005)

Damage to Host (Disease)

- **Superantigens**
 - Hyperstimulate the immune system
 - 1/5 T cells stimulated rather than 1/10,000
 - Fever, nausea, diarrhea, vomiting
 - Leads to shock
 - Organ failure, circulatory collapse
 - Cause of toxic shock syndrome (TSST)
 - *Staphylococcus aureus* and *Streptococcus pyogenes*



Bob Crimi

Although TSST-1 accounts for more than 90% of cases associated with menstruation, other enterotoxins account for 50% of cases unrelated to menstruation.

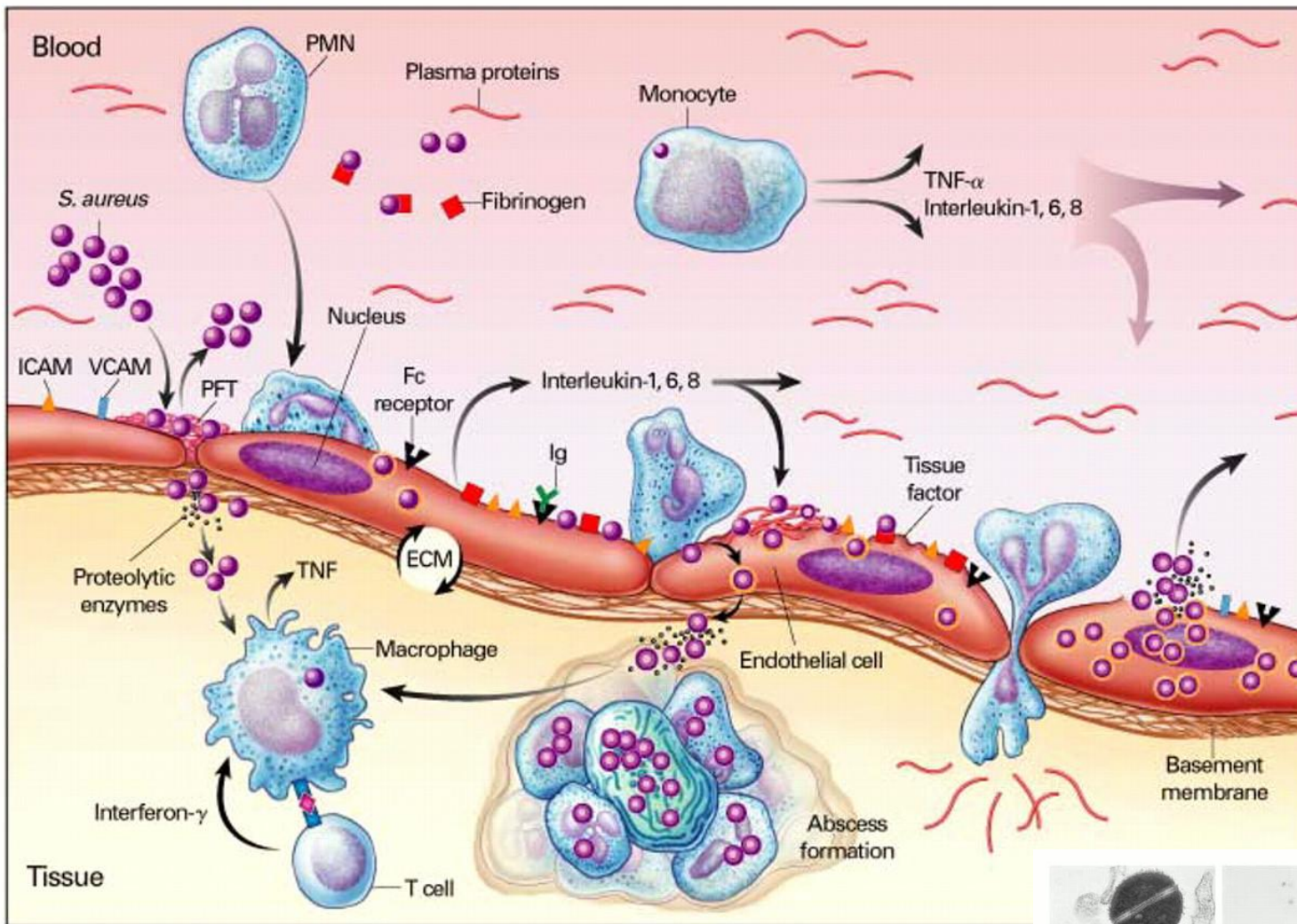
Nonmenstrual cases have increased and now account for approximately 1/3 of all cases.

Nonmenstrual cases have been associated with surgery, localized infections, and insect bites.

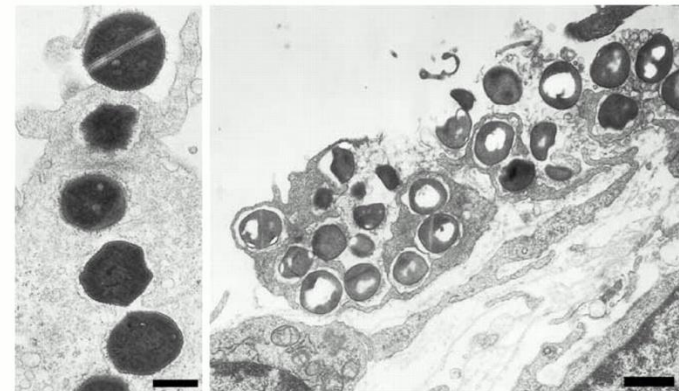
Nonmenstrual case have a higher mortality rate.

S. aureus Sepsis

- Minority of bacteremic or local infections progress to sepsis.
- Risk factors include advanced age, immunosuppression, chemotherapy, and invasive procedures.
- Presents similar to Gram-negative sepsis: fever, hypotension, tachycardia, and tachypnea.
- *S. aureus* one of the most common Gram-positive pathogens in sepsis cases.
 - levels of IL-6, IL-1, and TNF- α are predictive of outcome.



Origins of Sepsis



S. aureus Endocarditis

- Staphylococci cause 20-30% endocarditis.
- 80-90% are coagulase positive.
- Staphylococci attack normal valves.
 - important complication following valve replacement.
- Produce bulky vegetations.

S. aureus Bone Infections

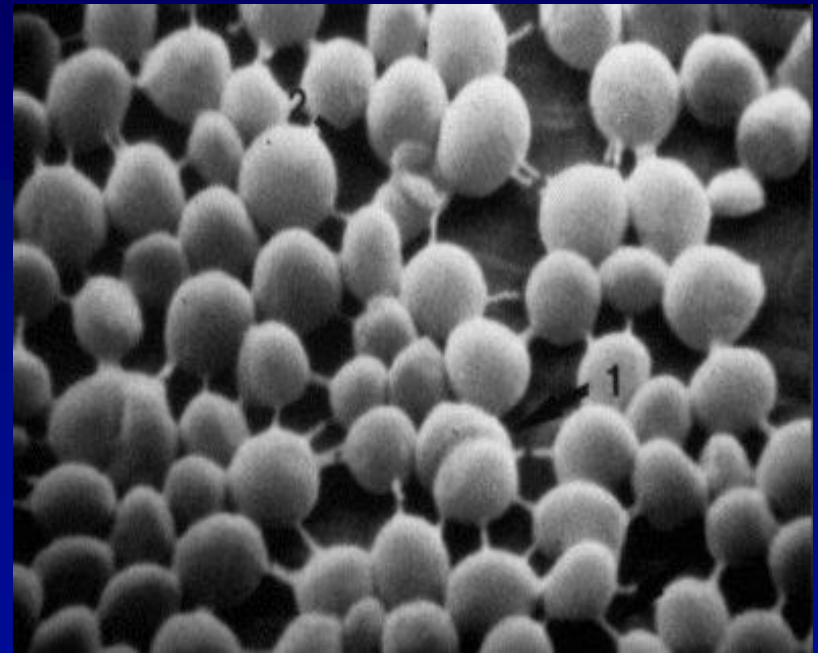
- Staphylococci are the most common cause of osteomyelitis, accounting for about half of all infections.

S. aureus Food Poisoning

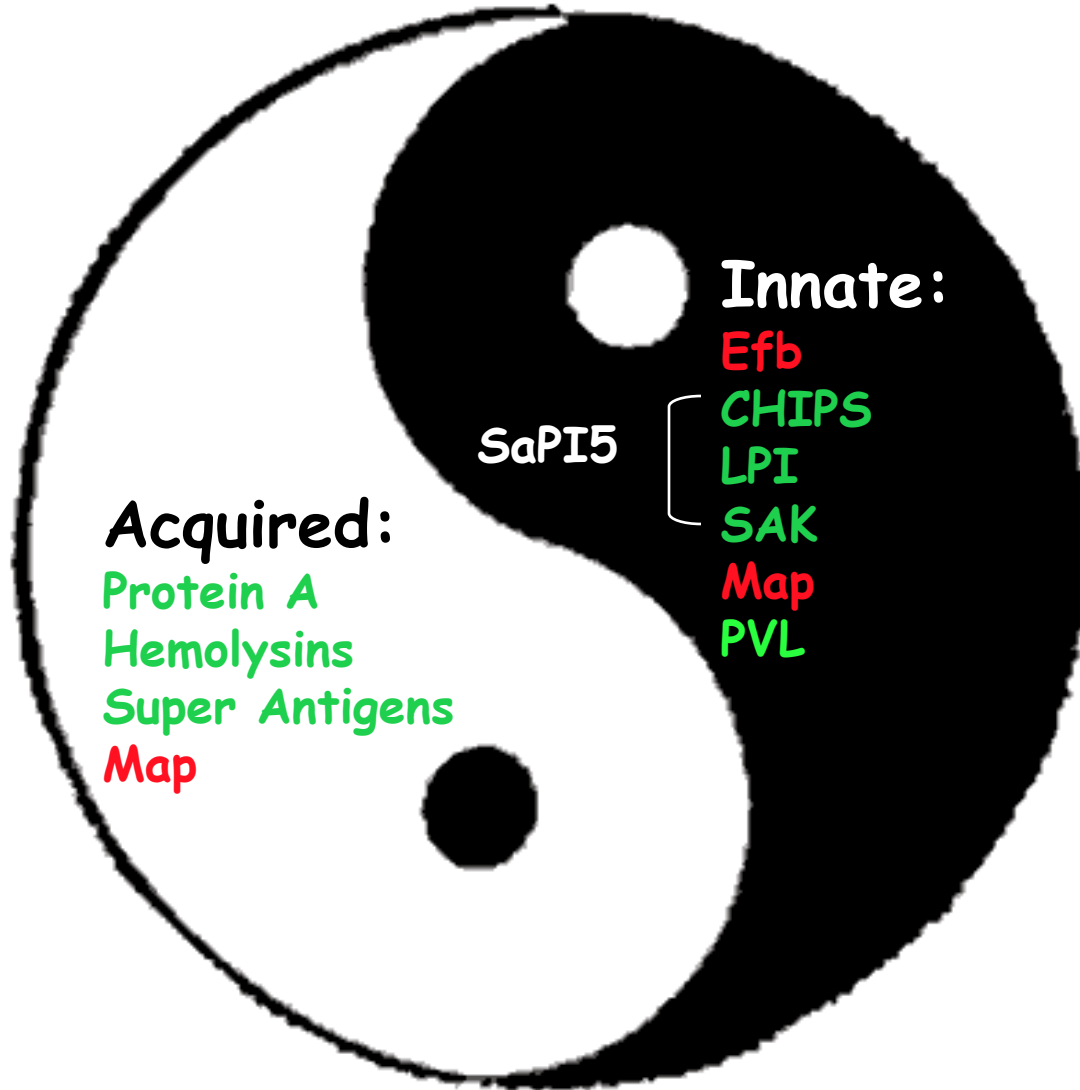
- Not an infection but a toxemia.
- Caused by one of several **heat-stable** enterotoxins (A-F).
- About 1/2 of all isolates produce one of these enterotoxins.
- Foodpoisoning caused by ingesting toxin-producing *S. aureus*.
- Vomiting begins within a few hours.
 - occasional diarrhea without blood or pus.
 - recovery within 24-48h
 - mechanism of action unknown

Staphylococcus aureus

- Individuals infected with SA do not usually develop protective immunity and are susceptible to reinfection.
- Suggested that SA can affect immune function



SA MIMs...Virulence Factors



Protein A, *spa*

- ~42 kDa
- Cell-wall associated and secreted forms
- Binds the Fc region of IgG
- Variable repeat number
- B-cell super antigen

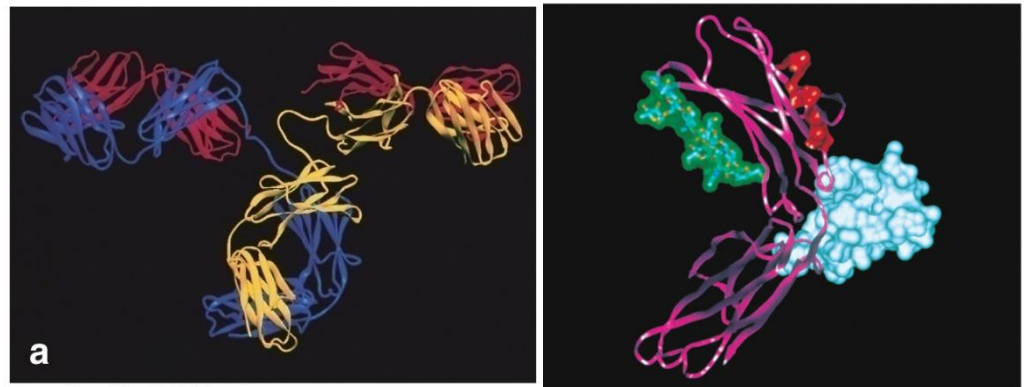
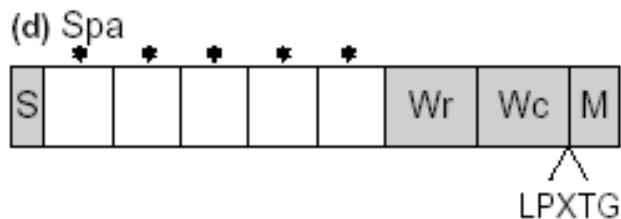


Figure 3-1 part 1 of 3 Immunobiology, 6/e. (© Garland Science 2005)

Figure 3-1 part 2 of 3 Immunobiology, 6/e. (© Garland Science 2005)

Staphylococcal toxins (Table 22-3)

- α -toxin-

- encoded by chromosome or plasmid.
- Found on most strains.
- Disrupts smooth muscle cell in blood vessels.
- Toxic to erythrocytes, leukocytes, hepatocytes, and platelets.

- β -toxin-

- also called sphingomyelinase C.
- Specificity for sphingomyelin.
- Toxic for erythrocytes, fibroblasts, leukocytes and macrophages.

- δ -toxin-

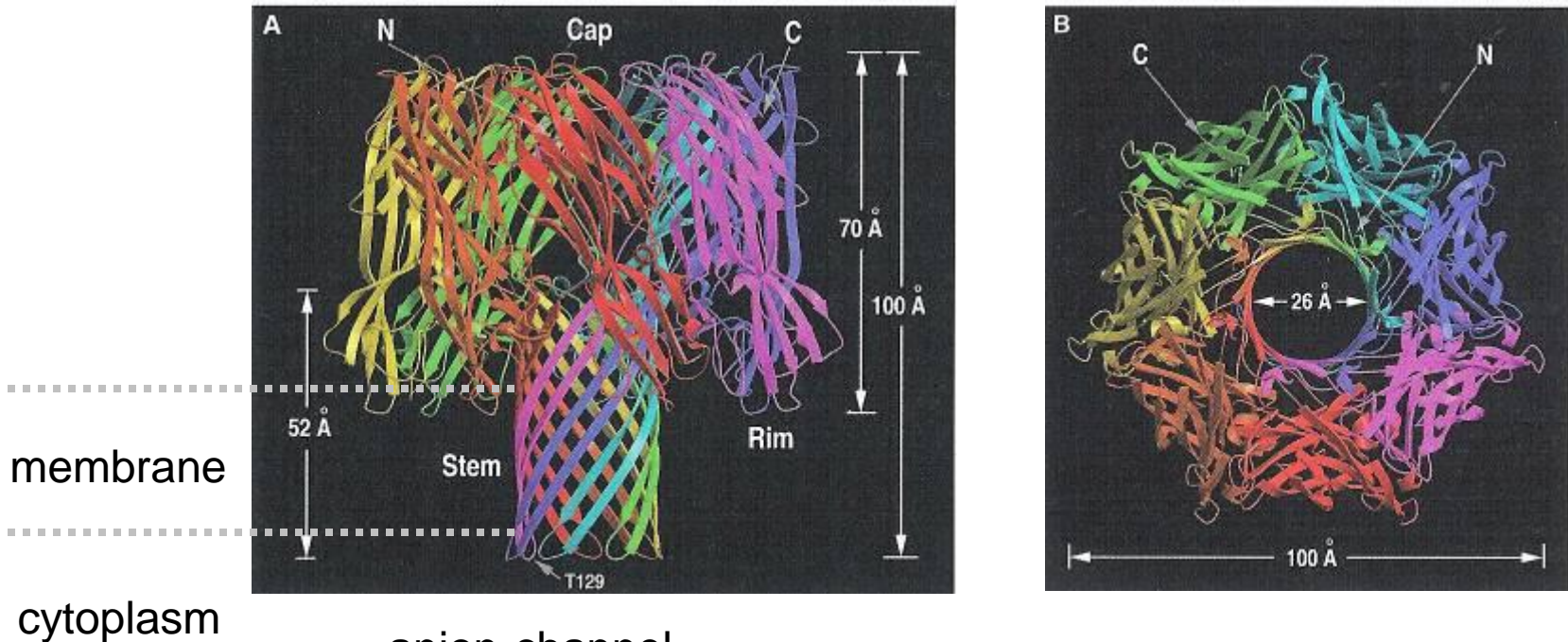
- wide spectrum of cytotoxicity. Less specific.

- γ -toxin-

- PVLs...

Structure of Staphylococcal α -Hemolysin, a Heptameric Transmembrane Pore

Langzhou Song,* Michael R. Hobaugh,* Christopher Shustak,
Stephen Cheley, Hagan Bayley, J. Eric Gouaux†
Science (1996), Vol. 274, pp. 1859-1866

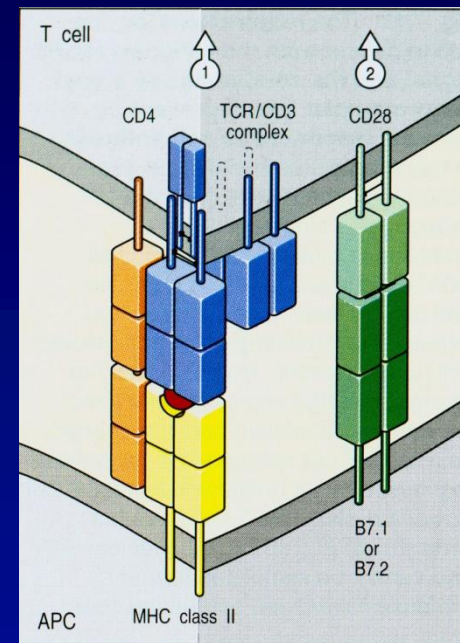
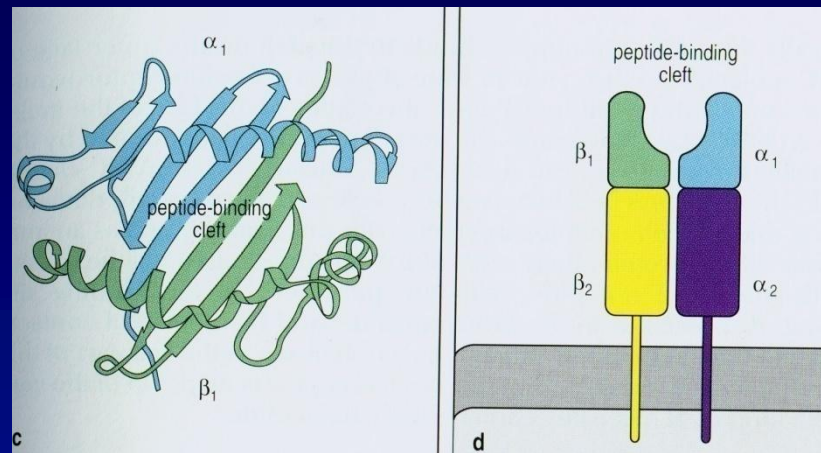


anion-channel
permeable for anions up to 0.6 nm diameter
however: mechanism of oligomerization/pore
formation in host cell membrane still unclear

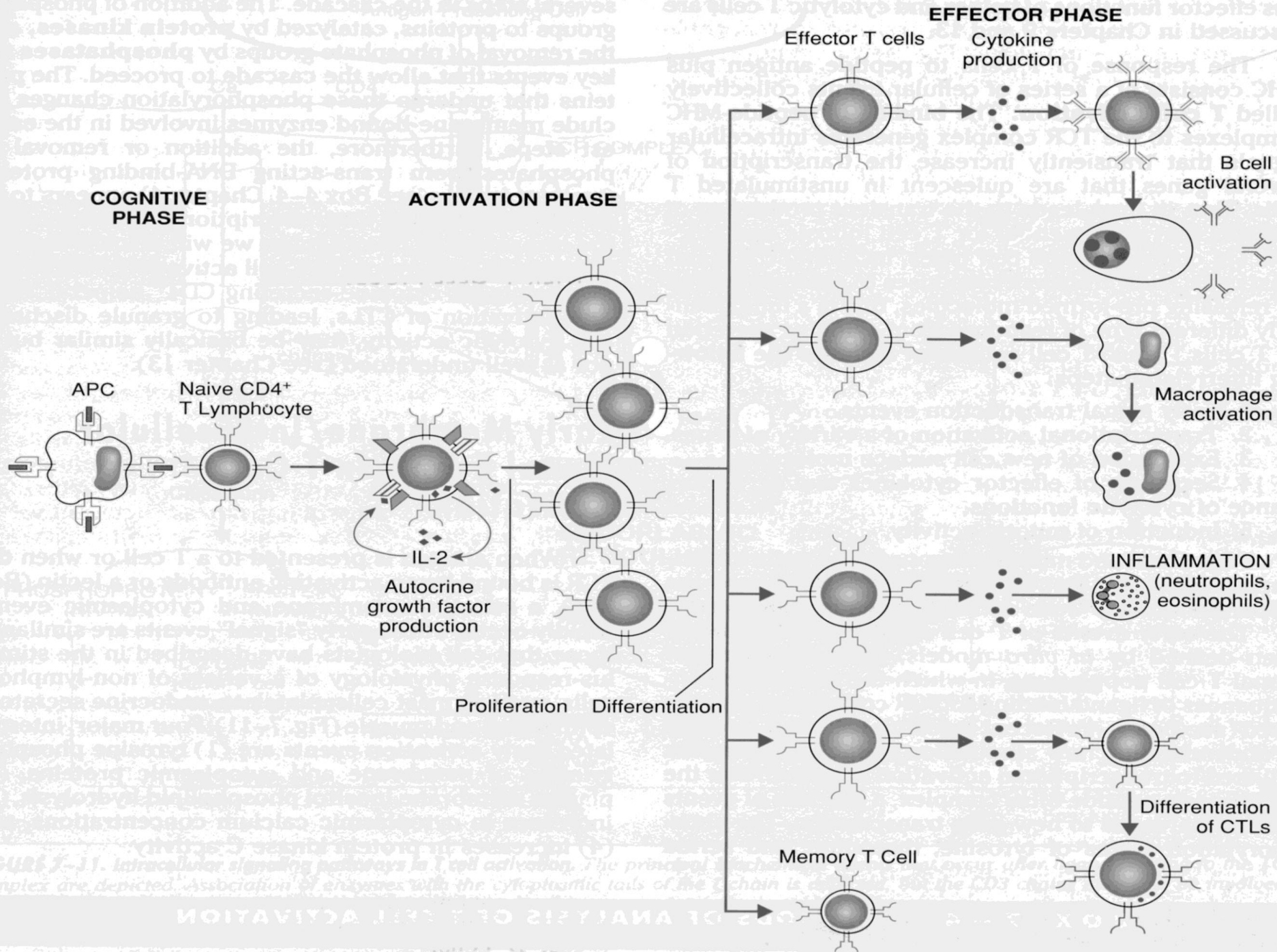
MHC class II Analog Protein (Map)

- 1st identified in *S. aureus* FDA 574 (McGavin, Jönsson)
- Composed of 689 A.A., 72 kDa
- Secreted
- MSCRAMM family
- Binds ECM (BSP, FN, TSP, VN, FBG, Op)
- MIM?

Major Histocompatibility Complex (MHC) class II protein



- Humans HLA
- Heterodimer (α/β)
- Expressed on M ϕ , B, Dc, and TE. \uparrow IFN- γ
- Processing and presentation of peptide Ag to CD4+ T cells.
- T cell activation and proliferation



Primary Structure of Map

| | | | | | | | |
|---|---|-----|-----|-----|-----|-----|-----|
| S | U | 1 | 2 | 3 | 4 | 5 | 6 |
| | | 48% | 48% | 29% | 28% | 29% | 10% |
| | | 61% | 65% | 52% | 59% | 52% | 59% |

Map 10

Map 19

- 6 domains of 110 A.A., 6 sub-domains
- A.A. sequence homology to peptide binding groove of MHC class II β

S. aureus FDA 574 Map

subdomain 1
subdomain 2
subdomain 3
subdomain 4
subdomain 5
subdomain 6

D-I-D-L-R-L-S-K-Q-A-K-Y-T-V-H-F-K-N-G-T-K-R-V-V-D-L-K-A-G-I-Y
D-V-D-L-R-L-S-K-Q-A-K-F-T-V-N-F-K-N-G-T-K-K-V-I-D-L-K-A-G-I-Y
E-R-D-L-K-H-A-K-K-A-Y-Y-T-V-Y-F-K-N-G-G-K-R-V-I-H-L-N-S-N-I-Y
G-E-R-E-L-K-Y-A-E-K-A-T-Y-T-V-H-F-K-N-G-T-K-K-V-I-N-L-N-S-N-I-S
D-I-E-L-K-F-A-K-Q-A-K-Y-T-I-H-F-K-N-G-K-T-Q-V-V-D-L-K-S-D-I-F
K-H-D-I-G-L-S-E-R-T-V-Y-K-V-Y-F-K-D-G-S-S-K-L-E-D-L-K-A-A-K-Q

MHC II

Human

HLA-DR-9a beta
HLA-DR-7 beta-1
HLA-DR2 Dw12a beta
HLA-DR2 AZHa beta
HLA-DR-2a beta
HLA-DR-1 beta
HLA-DQ-beta-1

G-D-T-Q-P-R-F-L-K-Q-D-K-F-E-C-H-F-F-N-G-T-E-R-V-R-Y-L-H-R-G-I-Y
G-D-T-Q-P-R-F-L-E-Q-A-K-C-E-C-H-F-L-N-G-T-E-R-V-W-N-L-I-R-Y-I-Y
G-D-T-R-P-R-F-L-Q-Q-D-K-Y-E-C-H-F-F-N-G-T-E-R-V-R-F-L-H-R-G-I-Y
G-D-T-R-P-R-F-L-Q-Q-D-K-Y-E-C-H-F-F-N-G-T-E-R-V-R-F-L-H-R-G-I-Y
G-D-T-R-P-R-F-L-Q-Q-Q-K-Y-E-C-H-F-F-N-G-T-E-R-V-R-F-L-H-R-D-I-Y
G-D-T-R-P-R-F-L-W-Q-L-K-F-E-C-H-F-F-N-G-T-E-R-V-R-L-L-E-R-C-I-Y
R-D-S-P-E-D-F-V-Y-Q-F-K-G-L-C-Y-F-T-N-G-T-E-R-V-R-G-V-T-R-H-I-Y

Mouse

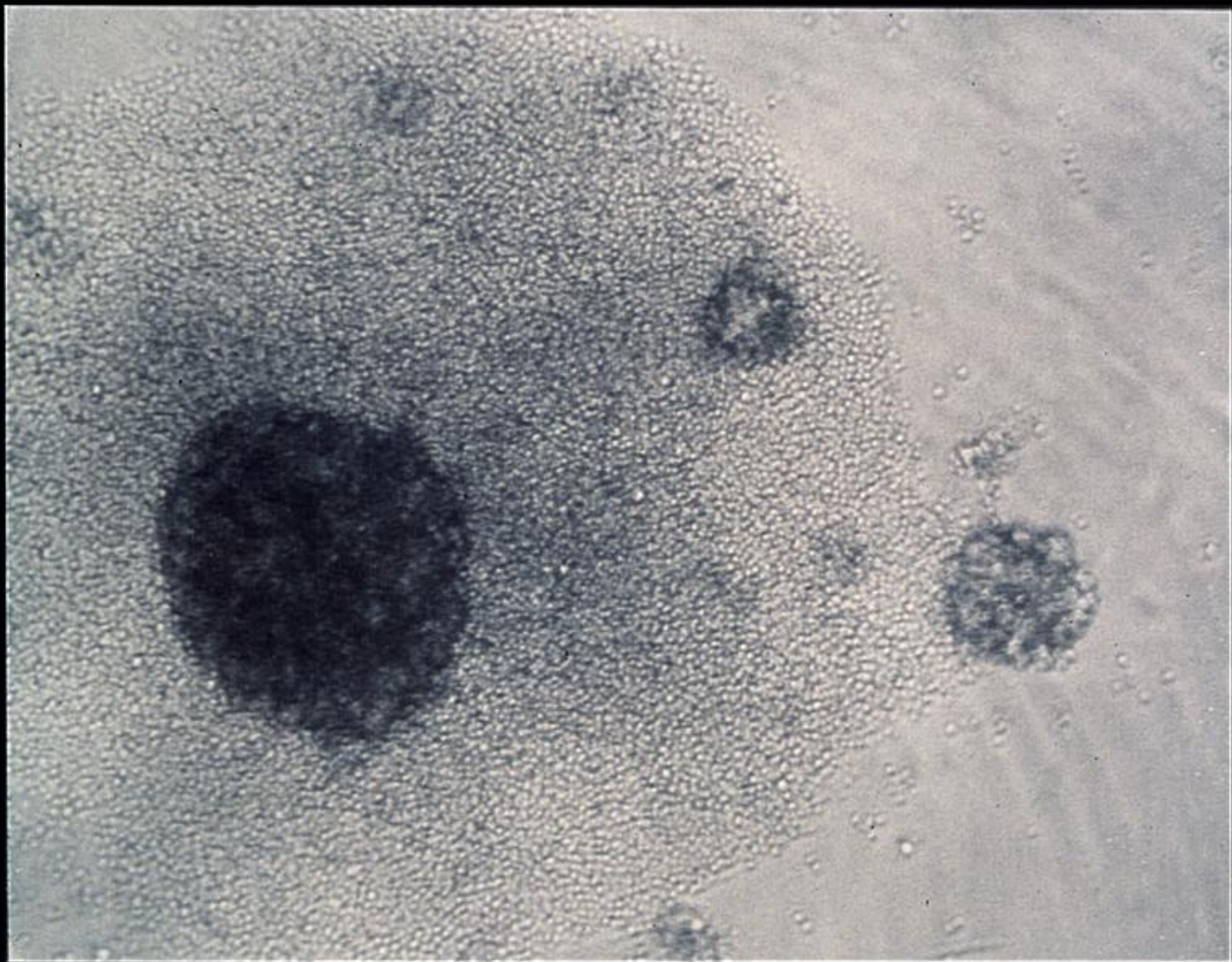
I-E beta
E-D beta
I-A beta
A-S beta
E-1 beta

R-D-S-E-P-W-F-L-E-Y-C-K-S-E-C-H-F-Y-N-G-T-Q-R-V-R-L-L-E-R-Y-F-Y
R-D-S-R-P-R-F-L-E-Y-V-T-S-E-C-H-F-Y-N-G-T-Q-H-V-R-F-L-E-R-F-I-Y
G-D-S-E-R-H-F-V-Y-Q-F-M-G-E-C-Y-F-T-N-G-T-Q-R-I-R-Y-V-T-R-Y-I-Y
G-D-S-E-R-H-F-V-F-Q-F-K-G-E-C-Y-F-T-N-G-T-Q-R-I-R-Y-V-T-R-Y-I-Y
R-F-L-E-Q-A-K-H-E-C-H-F-Y-N-G-T-Q-R-V-R-F-L-L-R-Q-I-H

Monkey

DR-beta-5-0304
DR-beta-5-0101

K-Q-E-K-Y-E-C-H-F-F-N-G-T-E-R-V-R-F-L-H-R-G-I-Y
K-Q-D-K-Y-E-C-H-F-F-N-G-T-E-R-V-R-F-L-H-R-G-I-Y





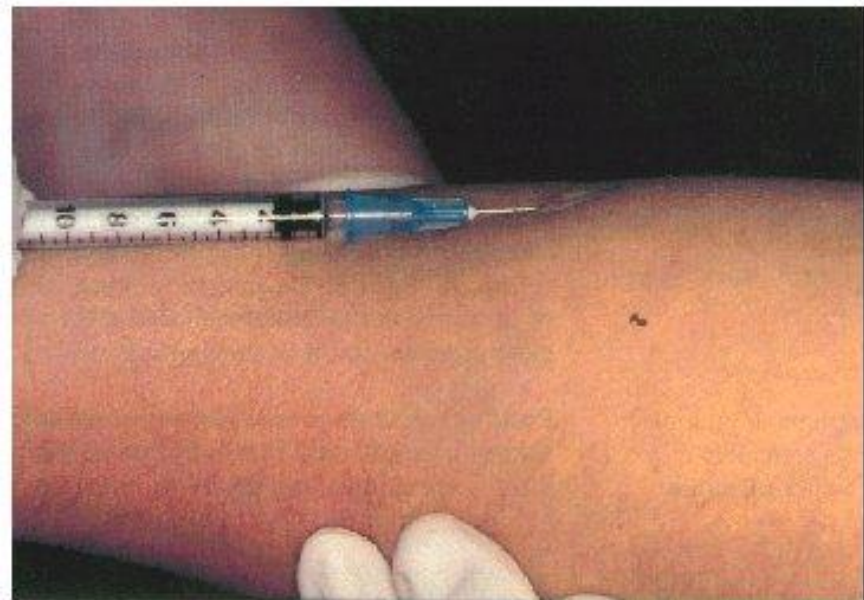
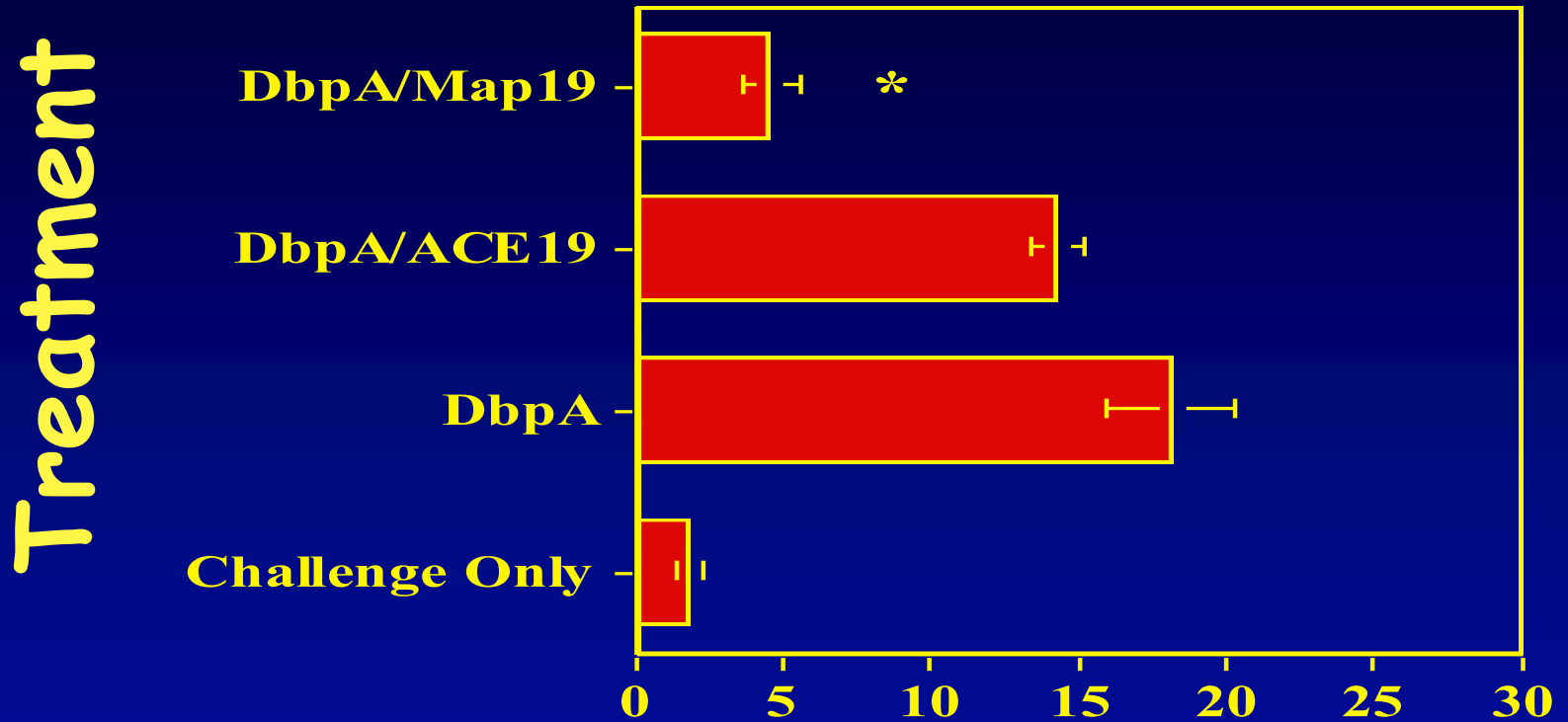


Figure 3.1 Giving the Mantoux tuberculin skin test.



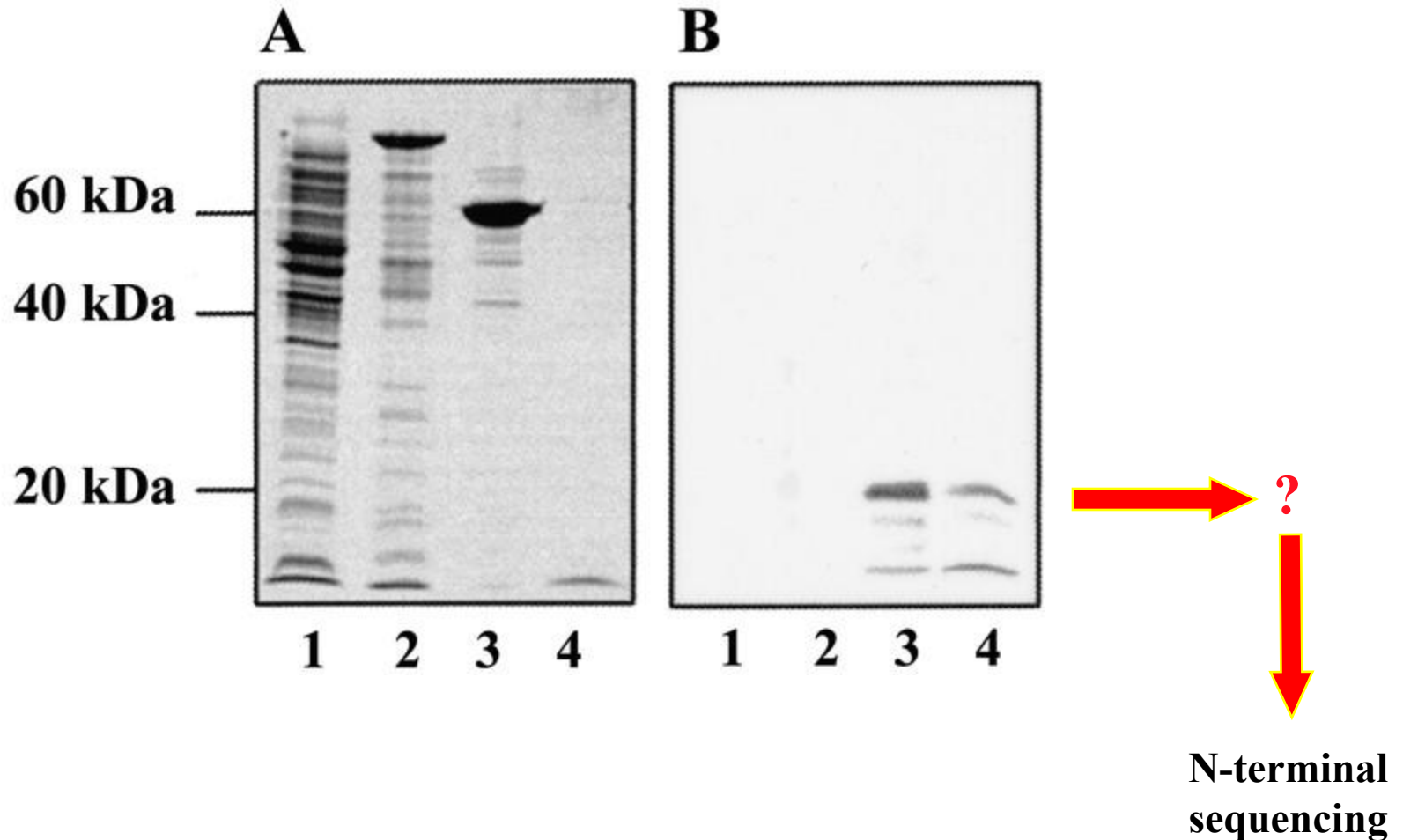
Effect of Map on DTH



Mean Footpad Swelling (0.01 mm)

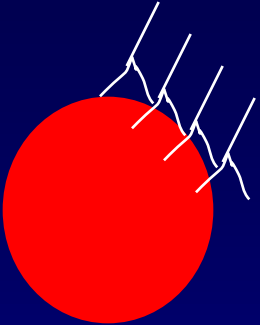
Can *S. aureus* bind C3?

SDS-PAGE and Western Ligand Blot of Whole Bacteria



Can Efb inhibit
complement activity?

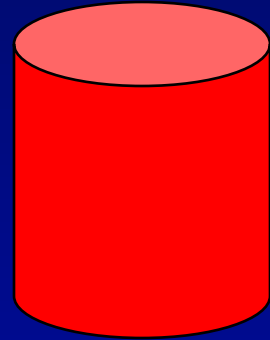
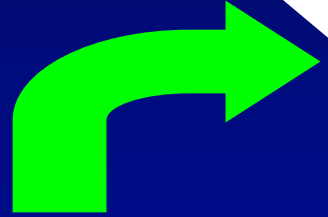
sRBC



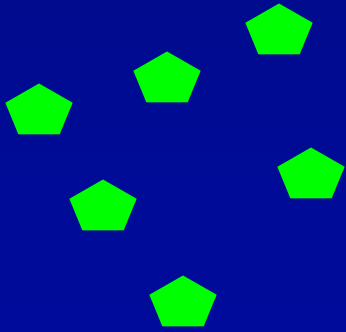
Antibodies



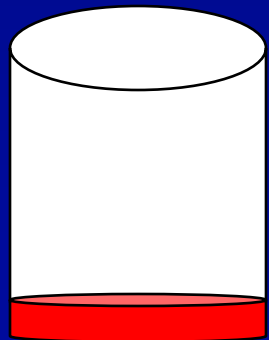
Serum/Complement



Lysis

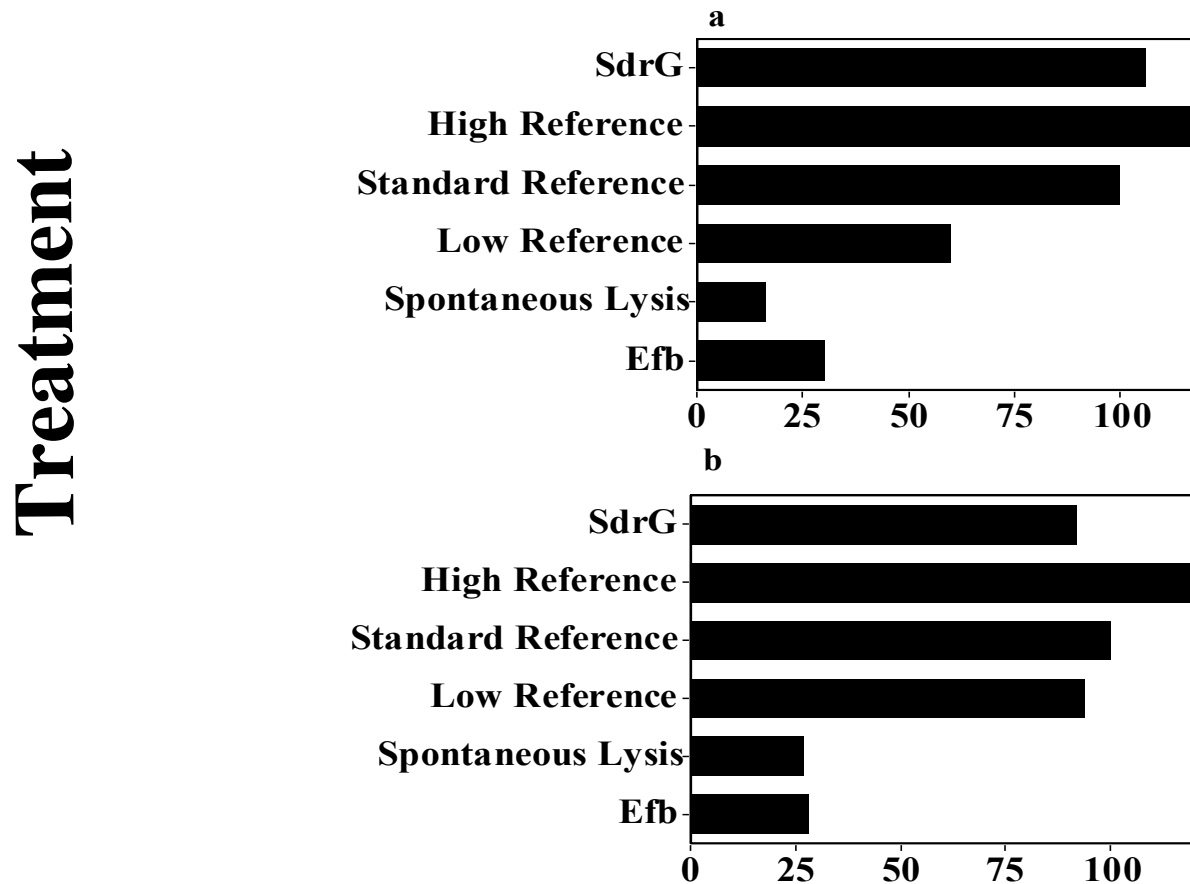


Efb



No Lysis

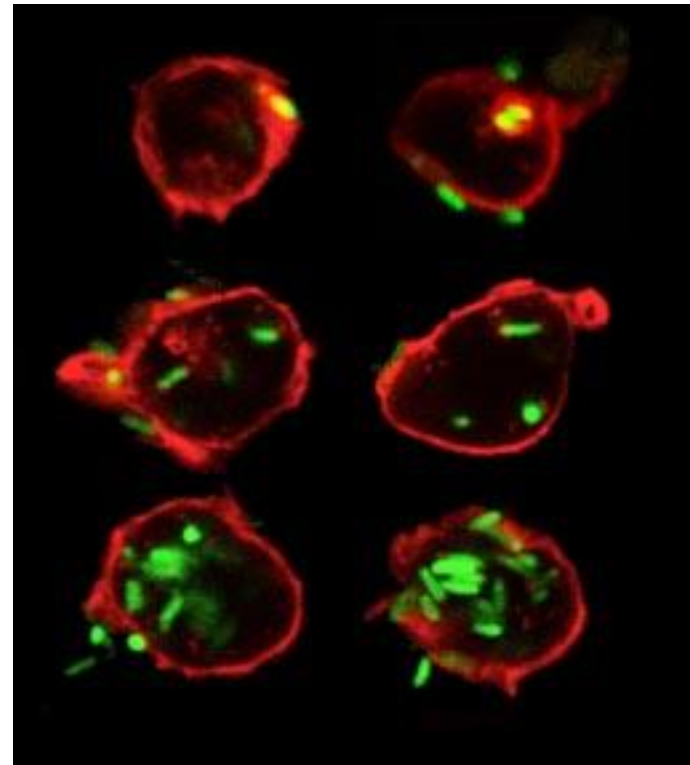
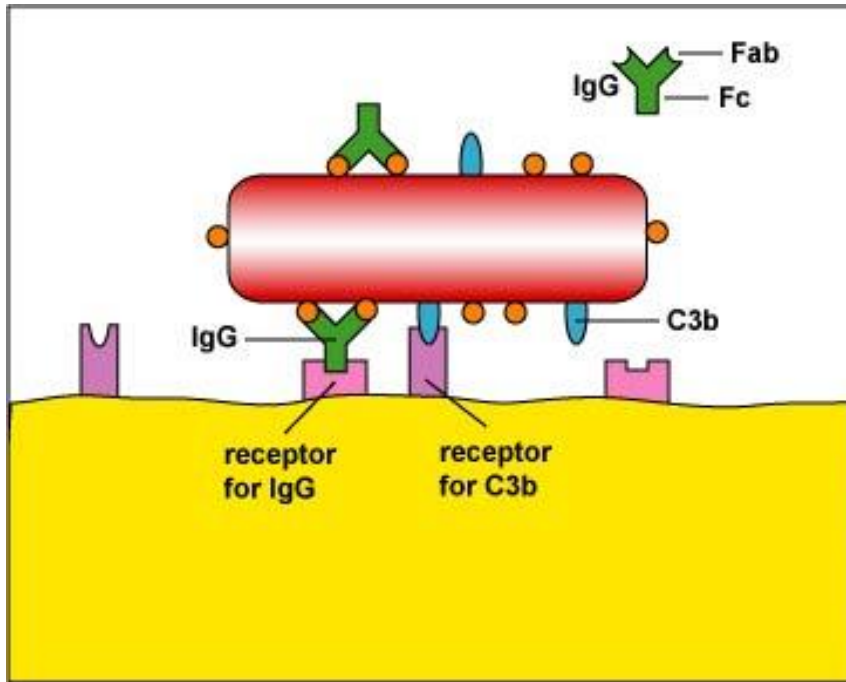
Complement-Mediated Lysis in the Presence of Various Proteins



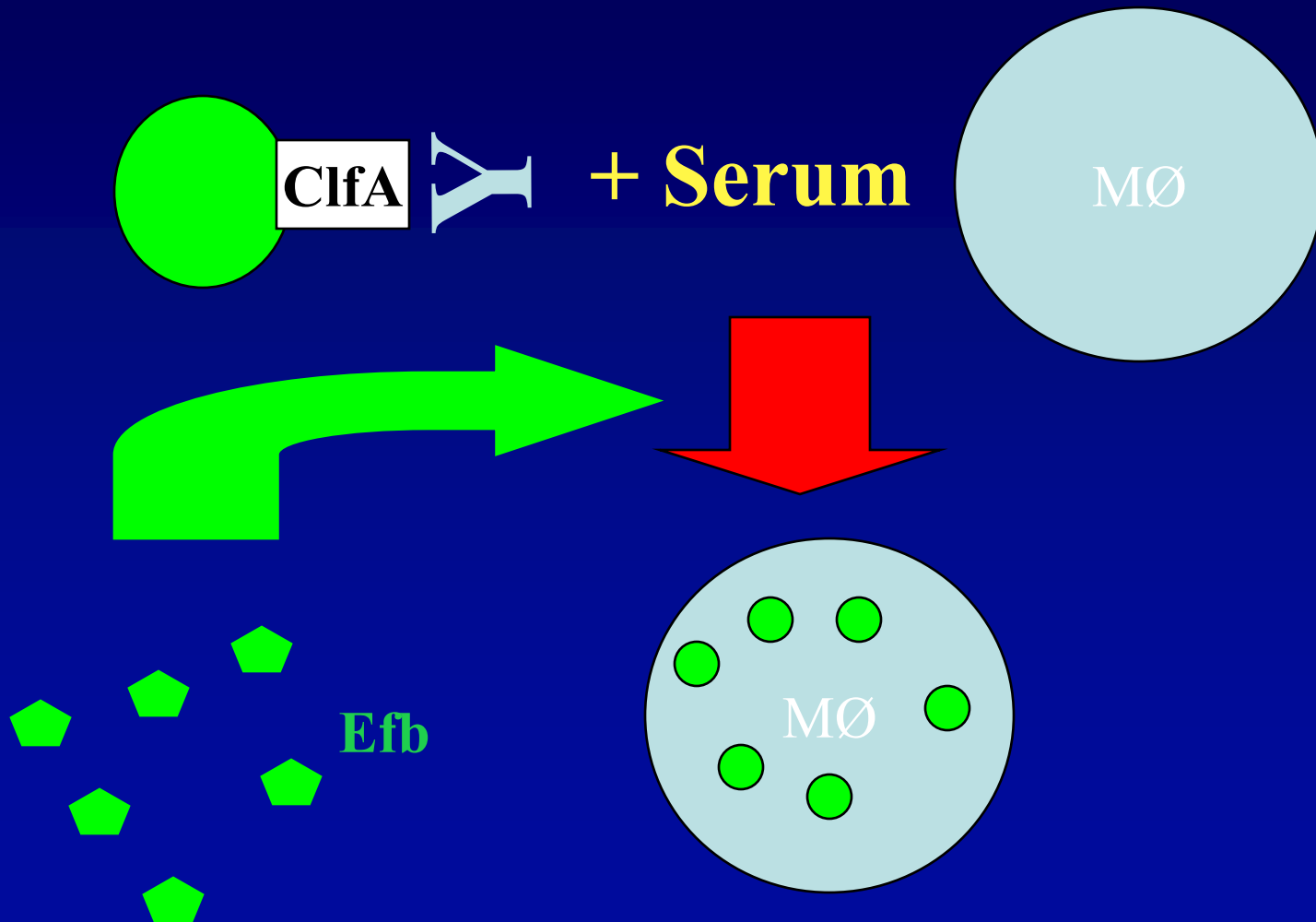
% of Complement Reference Serum

Can Efb Inhibit
Opsonophagocytosis?

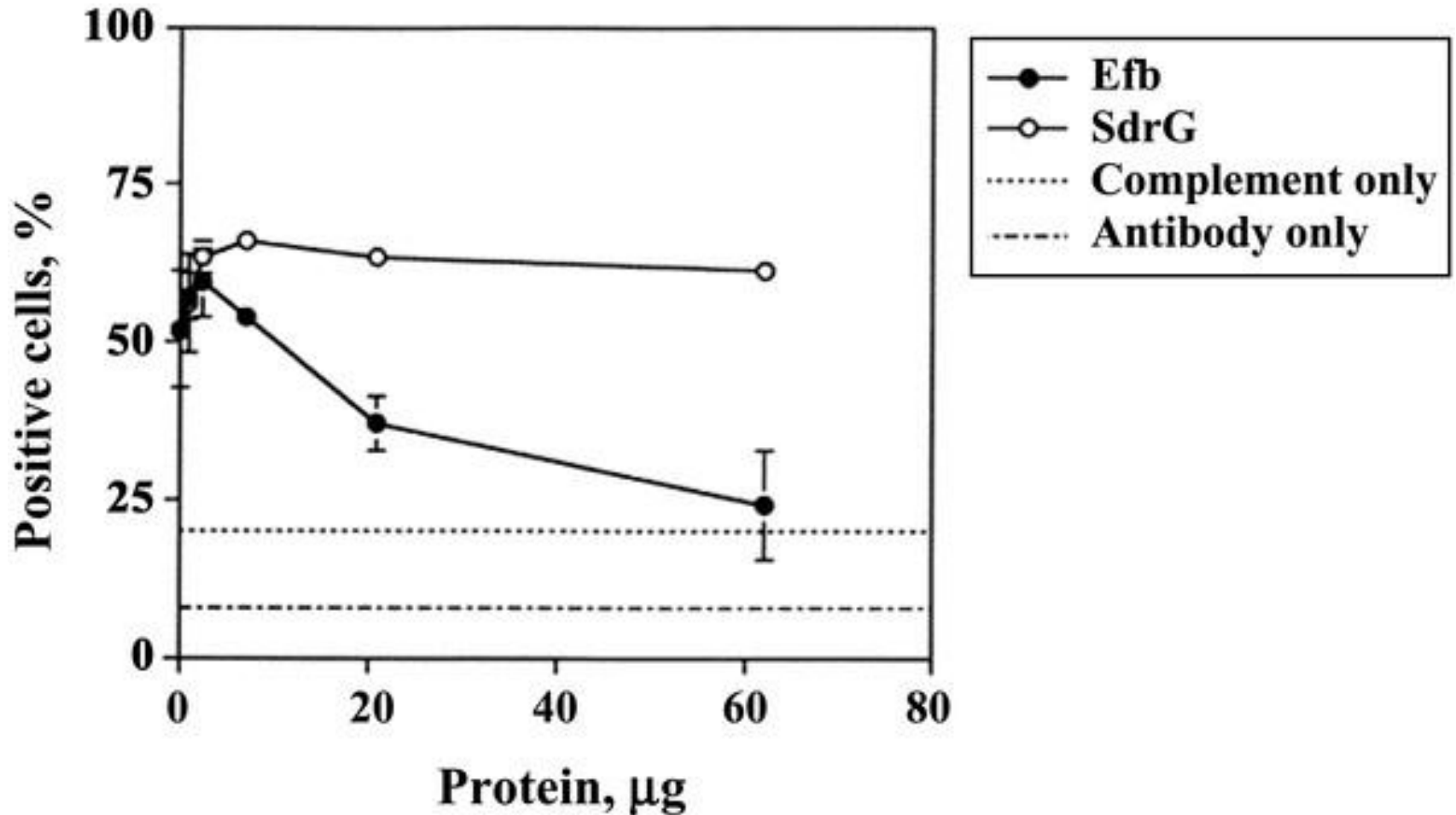
Opsonophagocytosis



Opsonophagocytosis of Fluorescent Beads



Efb Inhibits Complement-Mediated Opsonophagocytosis



Is the *Staphylococcus aureus* Panton-Valentine Leukocidin Associated with Disease Severity?

Community-Associated *S. aureus*



CA-MRSA: History of a Modern Emerging Pathogen

- "MRSA recently has been reported as an **outpatient pathogen**. We assessed this and found our institution's frequency quite low. Only 16 patients (4%) had true community-acquired MRSA bacteremia." *Infect. Control Hosp Epidemiol.* 19:106-107, 1998.
- "There is a low rate of bacteremia among patients **who have not been in contact with a health care facility** in the 6 month period prior to developing MRSA." *Clin. Infect. Dis.* 36:131-139, 2003.
- "In 2002, more than 900 people in Los Angeles county jails became infected with MRSA. Late last year, Los Angeles was the first city to record cases among gay men--at least 50 so far, says Ruane. Since then, the infection has cropped up in gay communities in cities such as San Francisco, Boston, Atlanta, and Washington, D.C., suggesting that **the microbe has found a new niche** in which it can travel fast." *Science* 299:1639-1641, 2003.

THE WALL STREET JOURNAL

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Silent Epidemic

Defying Treatment, A New, Virulent Bug Sparks Health Fears

Drug-Resistant Staph Kills
Quickly and Randomly;
Antibiotics' Worrying Toll

Simon Sparrow Wakes Crying

By MARILYN CHASE

In April 2004, Simon Sparrow was a robust toddler, 17 months old and just learning to feed himself. Then he caught a cold.

He awoke with a cry at his family's Chicago home. His parents took him to the University of Chicago Children's Hospital. Emergency-room doctors X-rayed his chest and chalked up his symptoms to a virus and asthma. They let him go home at about 1 p.m. At 4:30 p.m., his mother called back and asked doctors to listen to his worsening breathing over the phone.

Simon Sparrow

Call 911, they said. An ambulance whisked Simon back to the hospital where his condition



What's News—

Business and Finance

GOOGLE WILL "vigorously" oppose a Justice Department legal motion requiring it to disclose information about consumer Web searches. The standoff involves only anonymous data but follows cases where U.S. Internet firms have turned over personal data about a user or cut off an individual's Web site in China, explicitly or apparently in cooperation with the Chinese government.

(Article on Page A3)

■ **Home construction dropped 8.9%** and building permits declined 4.4% in December, in a fresh sign of a housing slowdown. Jobless claims fell to the lowest level in six years.

■ **Home Depot plans to cut store openings by nearly half** over five years and focus more heavily on its professional supply business.

(Articles on Pages A2 and A3)

■ **Nasdaq is exploring whether to make an offer for the London Stock Exchange**, signaling ambitions beyond the tech sector.

(Article on Page C3)

■ **Motorola's net soared 86%** on handset sales and the recovery of

World-Wide

■ **BIN LADEN SURFACED**, but officials see his threat as much-reduced.

In an audiotape on al-Jazeera that the CIA found authentic, al Qaeda's terror lord claimed strikes in the U.S. are planned and offered a "truce" on unspecified "fair" terms, which drew White House scorn. The administration seized on the tape as illustrating the need for tough antiterror tactics. Some saw similarities to al Qaeda's message preceding the London bombings, but others argue setbacks such as reported killings of four top terrorists in a Predator attack in Pakistan Friday have rendered the group unable to back up its bluster. (Column 4)

Gonzales laid out for lawmakers the administration legal justification for domestic wiretapping without court warrants. He is to testify before the Senate Feb. 6. Britain's Blair faced criticism. A leaked memo said advisers saw CIA "renditions" as illegal but may have looked the other way.

■ **Dec. 15 Iraq voting featured many irregularities and instances of fraud**, international monitors said without questioning the results, which may be announced today. Two Baghdad suicide bombings killed at least 21. Iraq claimed the U.S. may release female Iraqi hostages as the kidnappers of reporter Carroll demand, but the U.S. denied it. Kerry is visiting Baghdad.

For Oil-Rich Brides, Caviar, Crystal, And 1,000 Guests

Persian Gulf Sees a Boom

In Extravagant Weddings;
Almonds Coated in Gold

By YASMINE EL-RASHIDI

DUBAI, United Arab Emirates—When Noor Al-Roomi planned her son's recent wedding, she knew she was up against tough competition from many other big weddings. "Everyone wants to hold a more impressive wedding than the family before," says Ms. Al-Roomi.

She managed to hold her own. Arrangements included bringing in Beluga caviar from the Caspian, tablecloths from Switzerland, a cake chef from France and flowers from Holland. Guests received as favors gold-rimmed crystal bowls filled with Belgian chocolates.



"You don't want to have anything that is easily found on the local market. It has to be exclusive," she explains. The groom's family spent \$250,000 on the six-hour affair.

The Al-Roomi nuptials are part of a wedding industry that is resurgent in the Persian Gulf economic boom fed

Last Stand

An Ambitious Company's Move

With \$40 Million War C
Skillling Calls on Law
With Business Exper

Hiring a Sociology Profe

By JOHN R. EMSHWILLER

HOUSTON—Four years of irruptions and intense news coverage made Enron a synonym for frazzle. But when the trial of former executives Jeffrey Skillling and F. Lee Lay begins Jan. 30, defense lawyers make a bold argument: Everything the company did was legal.

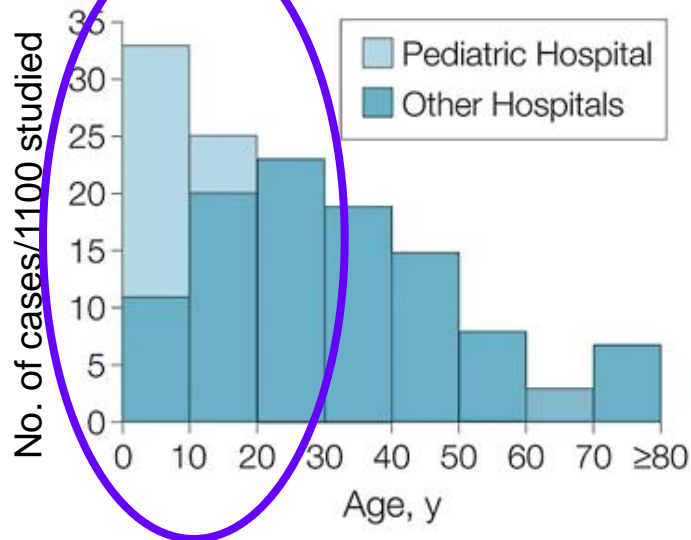
That approach stands in stark contrast to some other big-name co-defendants in recent history. Law firm former WorldCom Inc. Chief Executive Bernard Ebbers and former Health Corp. chief Richard Scrushy did dispute that large-scale financial shenanigans had occurred at the companies. They simply argued that they didn't know about the wrongdoing. New York jury convicted Mr. Lay while a Birmingham, Ala., jury acquitted Mr. Scrushy.

To make his case, Mr. Skillling's lawyer, Daniel Petrocelli, a civil litigator with no criminal-trial experience. The 50-year-old Mr. Petrocelli will argue that the government is trying to criminalize

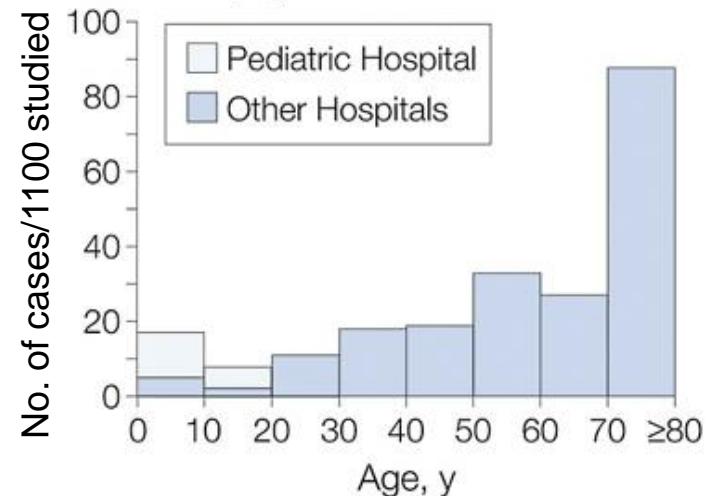
Community Associated MRSA (CA-MRSA)

- No previous MRSA history
- Affects healthy individuals
- Infection rates are higher in children and young adults

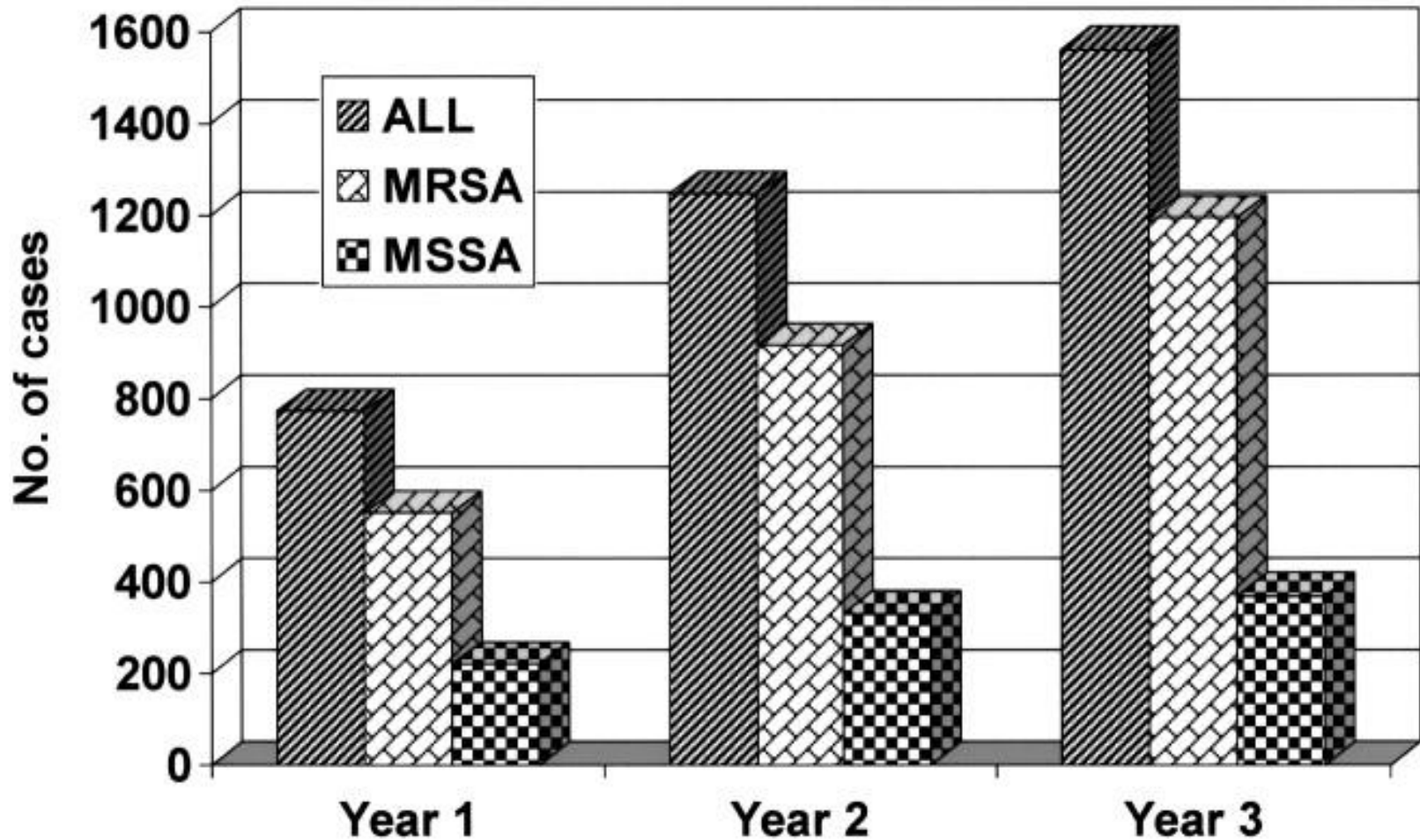
Community-Associated Methicillin-Resistant
Staphylococcus aureus



Health Care-Associated Methicillin-Resistant
Staphylococcus aureus

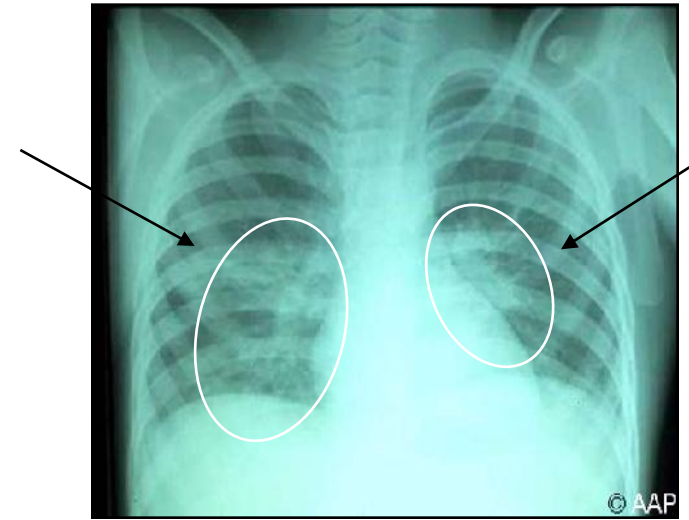


Community Associated *S. aureus* at Texas Children's Hospital



CA-MRSA cause invasive infections

- These strains cause severe infections:
 - Skin and muscle (79%)
 - Osteomyelitis (52%)
 - Pneumonia (21%)



CA-MRSA and Pneumonia

- Recently found to cause necrotizing pneumonia and death in healthy young people.
 - Associated with viral infection or preceded with influenza-like illness and CA-MRSA (Lancet 359: 753-759, 2002; Clin. Infect. Dis. 35:819-824, 2002)
- PVL: Pantone-Valentine Leukotoxin
 - Encoded by *lukPV* - bicomponent leukotoxin (like γ -toxin, LukE, D)
 - Carried by bacteriophage phiPVL and phiSLT.
 - In the past, PVL has been associated with severe skin infections
- Associated with CA-MRSA that are susceptible to antibiotics that are ineffective on nosocomial MRSA
- Associated with a specific *SSCmec* type

CA-MRSA: Necrotizing pneumonia

- 75% of patients die despite of antibiotic therapy
- 90% carry Panton Valentine Leukocidin (0.6% nasal carriage)

Association between *Staphylococcus aureus* strains carrying gene for Panton-Valentine leukocidin and highly lethal necrotising pneumonia in young immunocompetent patients

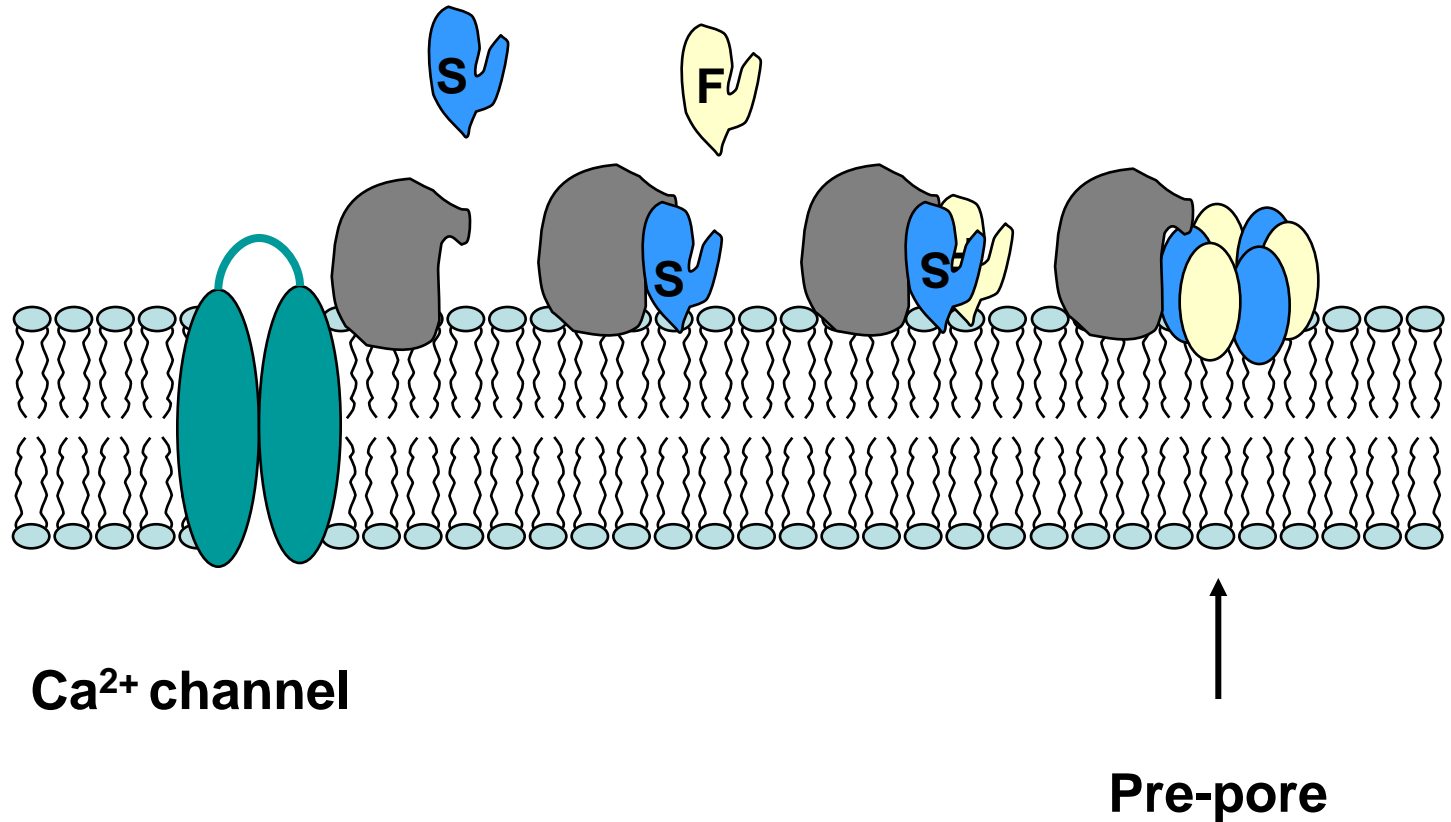
Yves Gillet, Bertrand Issartel, Philippe Vanhems, Jean-Christophe Fournet, Gerard Lina, Michèle Bes, François Vandenesch, Yves Piémont, Nicole Brousse, Daniel Floret, Jerome Etienne

Lancet 2002; 359: 753–59

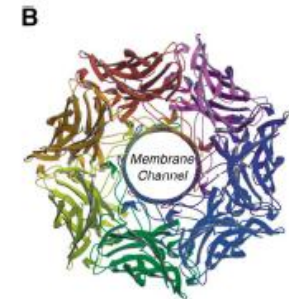
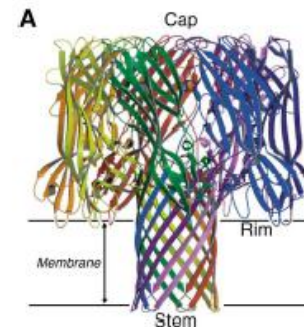
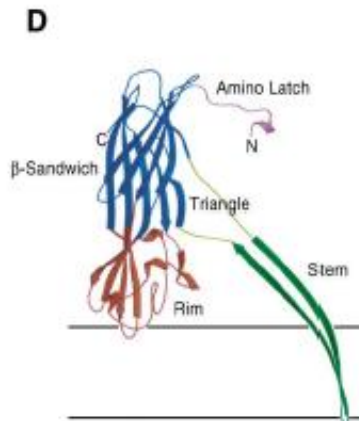
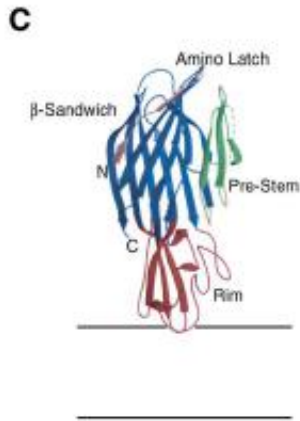
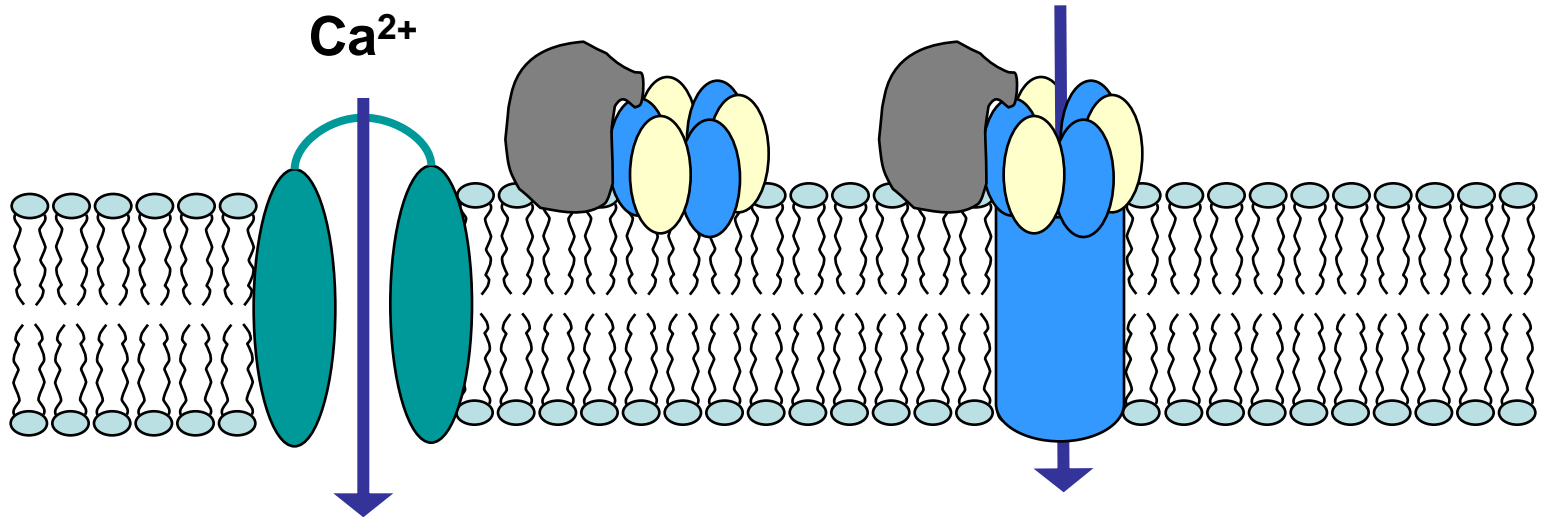
Panton Valentine Leukocidin

- Bi-component toxin.
- Mode of action is well known
 - create lytic pores
 - common targets: monocytes, macrophages and PMNs
- Encoded by *lukS-PV* and *lukF-PV* which are carried by bacteriophages.

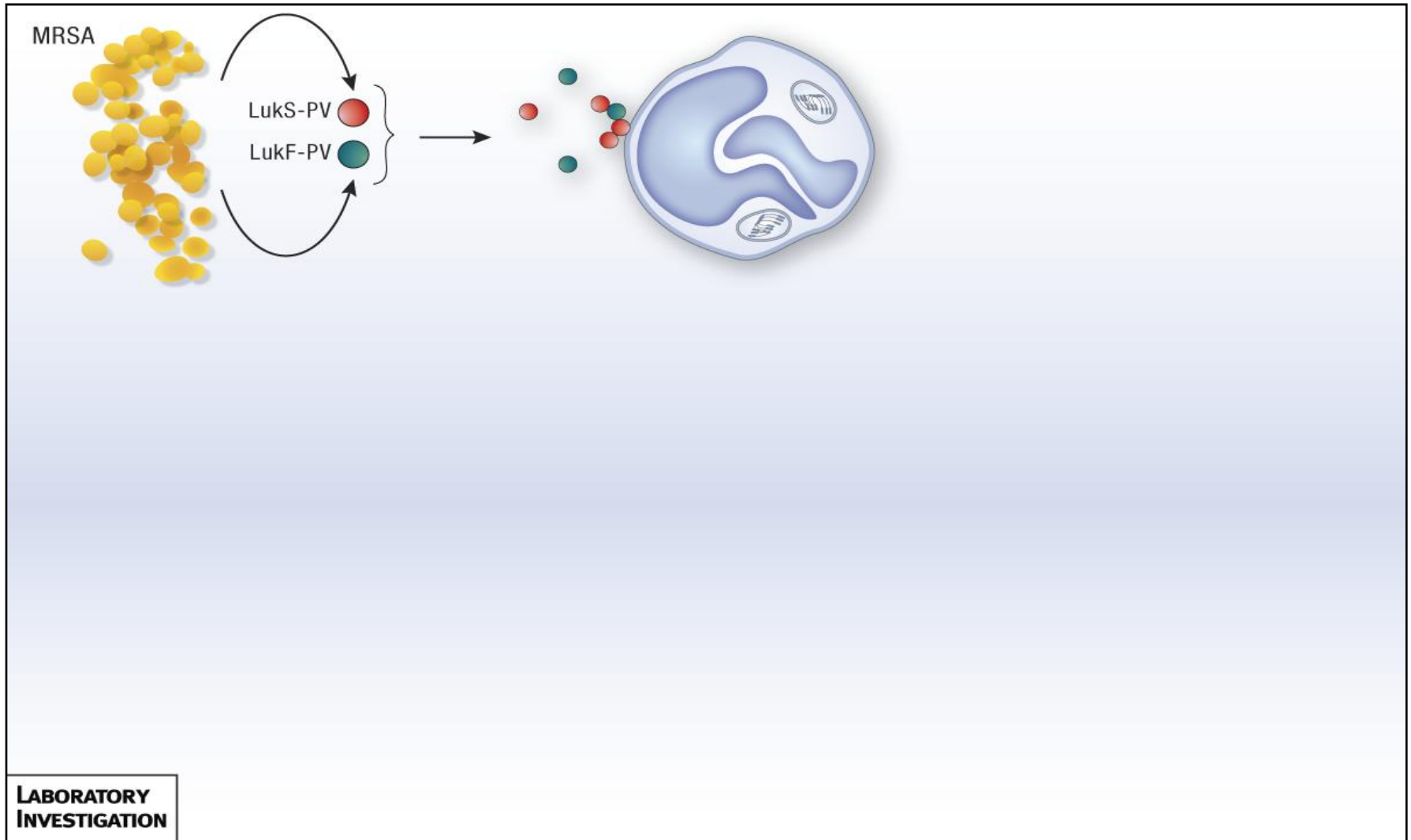
Mode of action: pre-pore



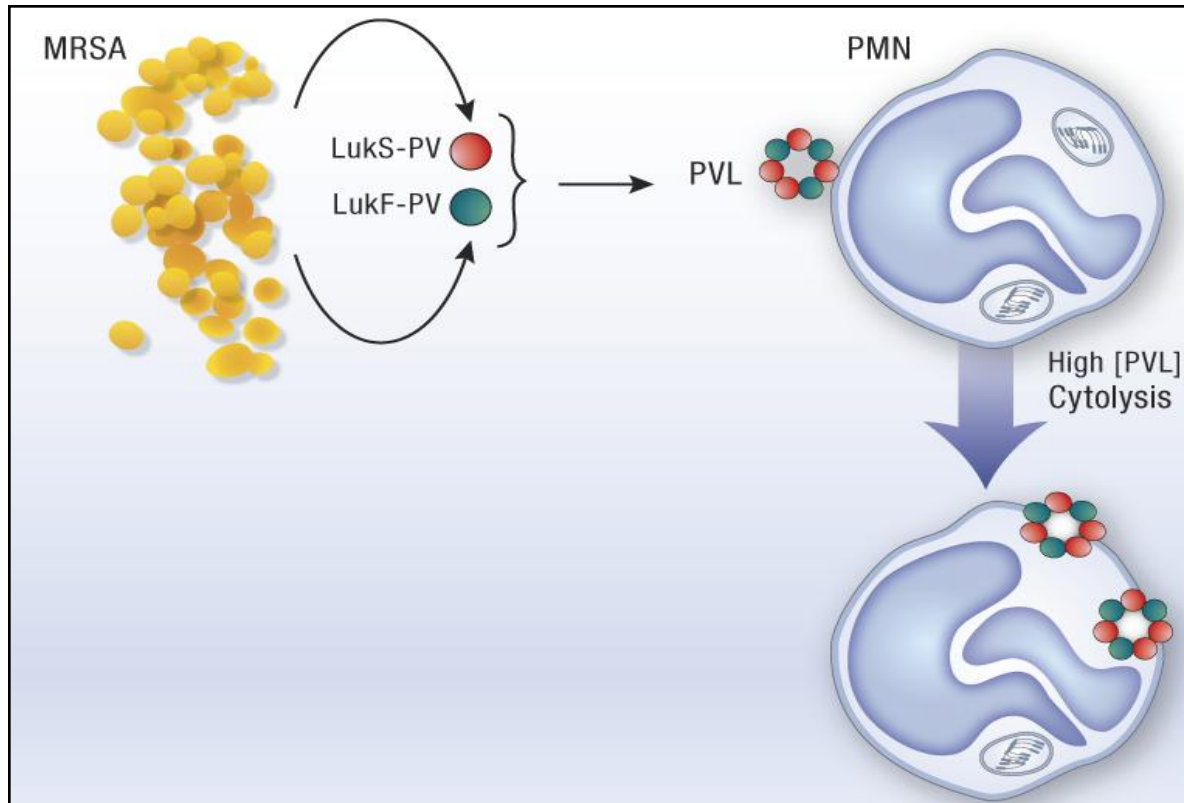
Mode of action: pore



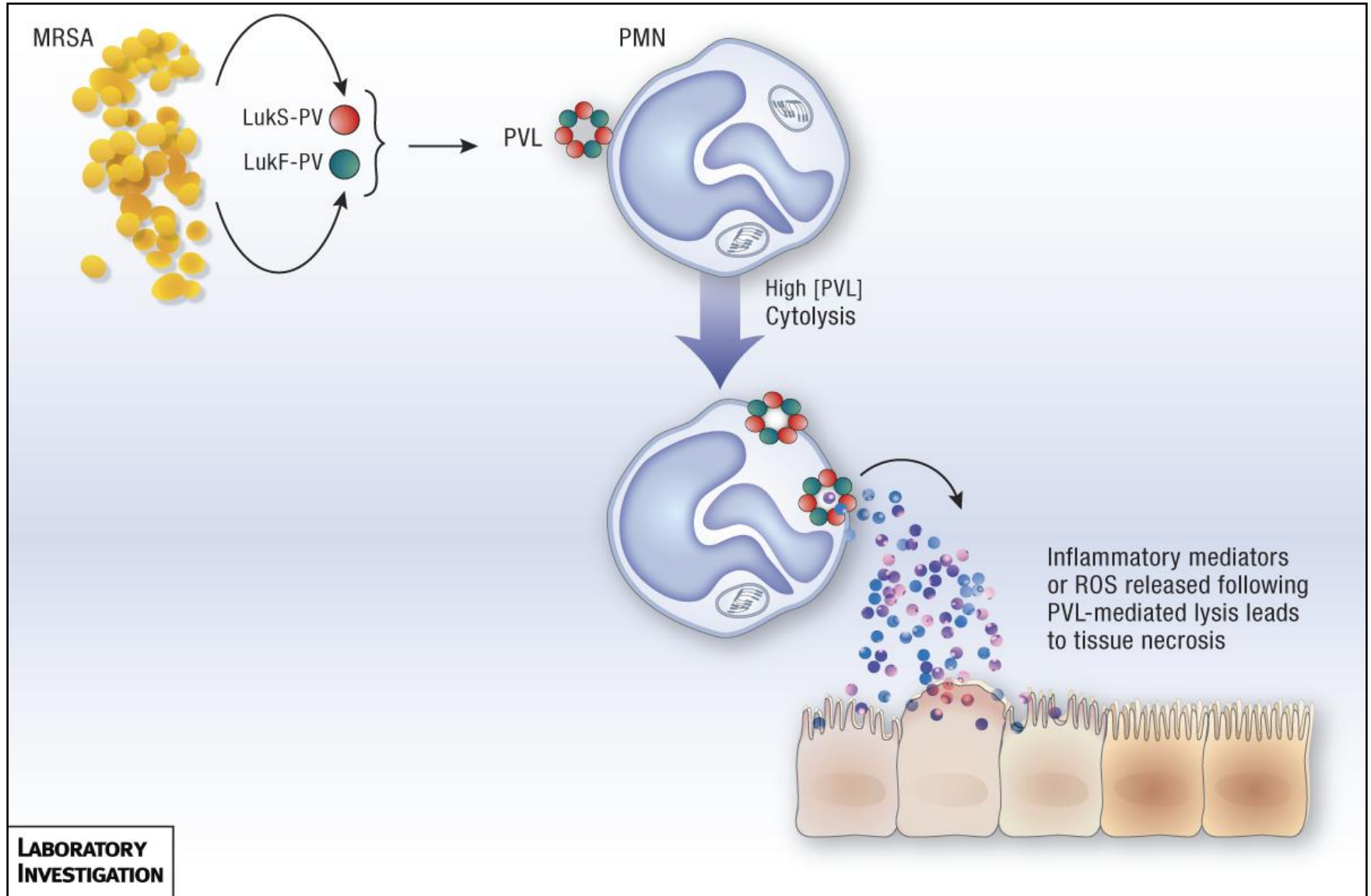
PVL secretion and assembly on PMN membrane



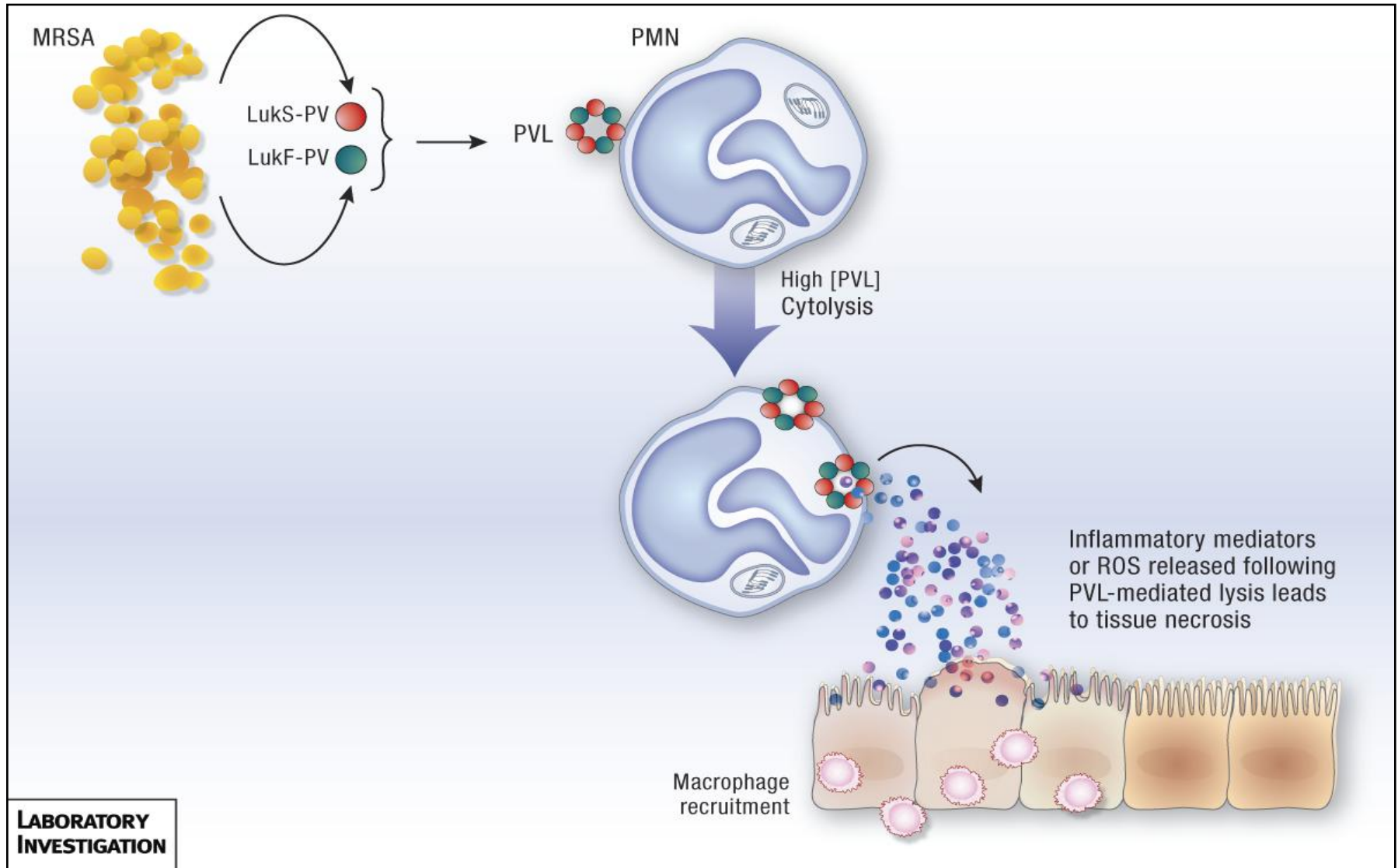
PMN cytolysis



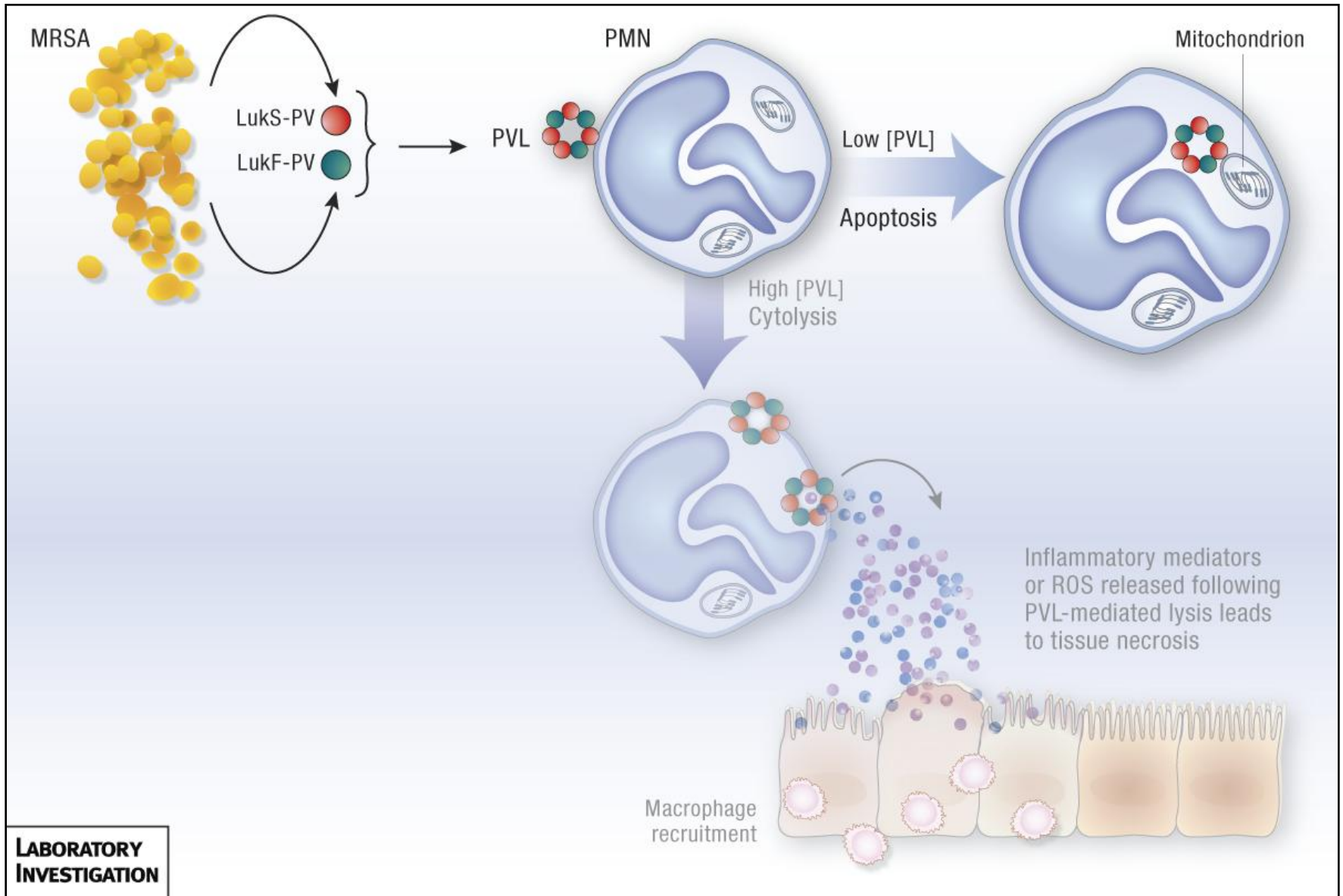
PVL-mediated lysis and tissue necrosis



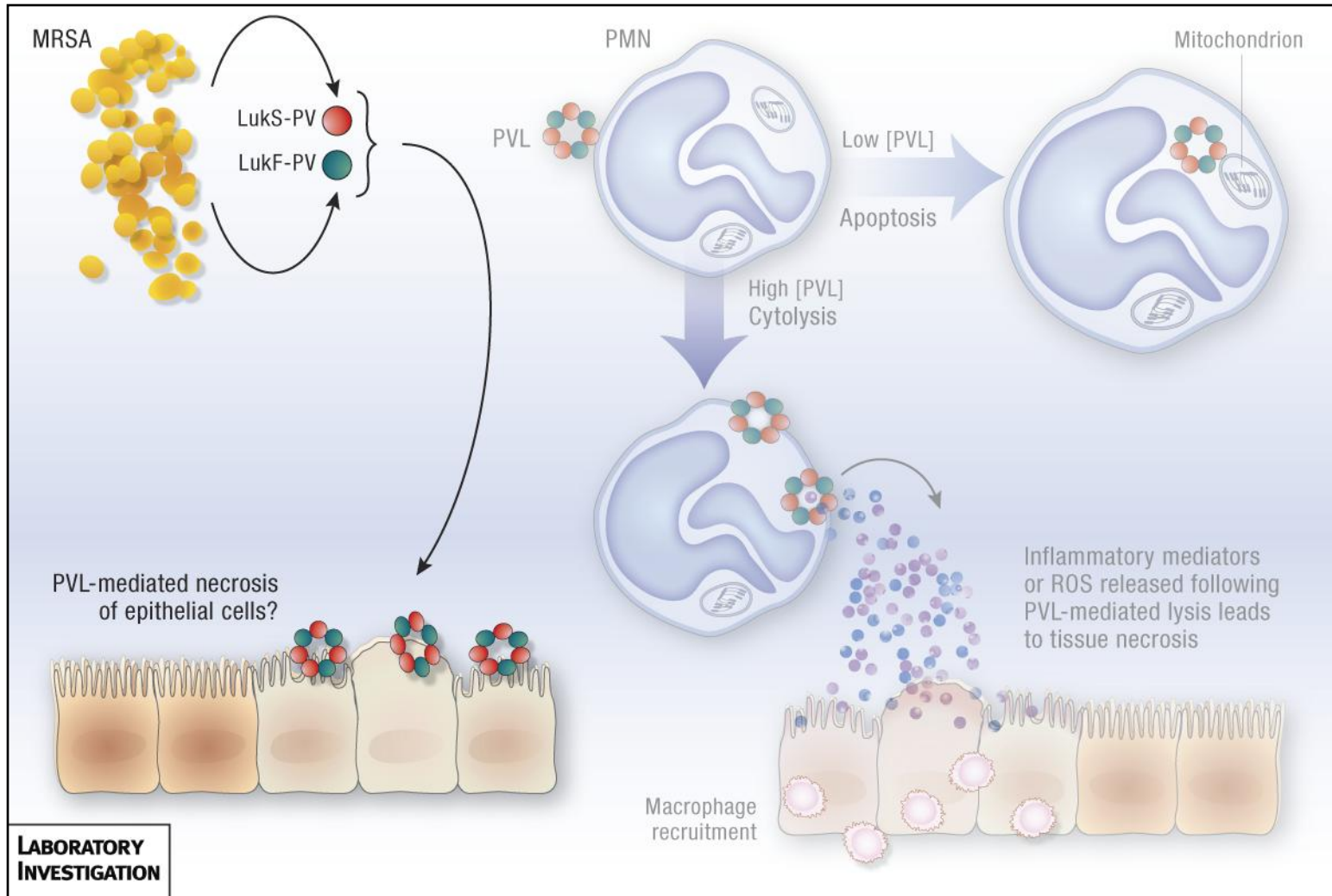
PVL-mediated lysis and tissue necrosis



PVL apoptosis



PVL-mediated necrosis?

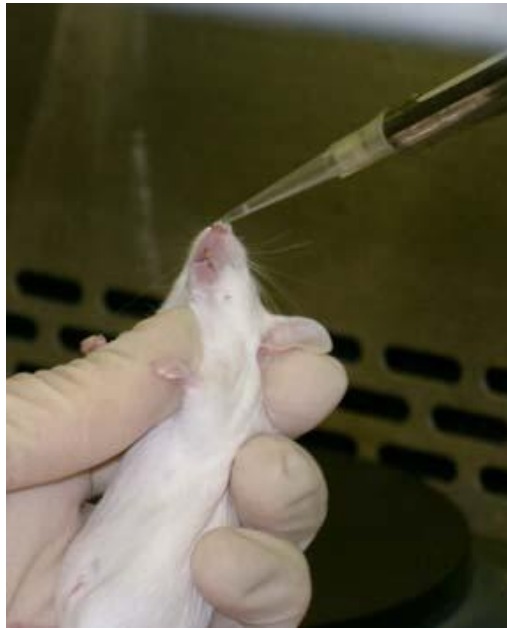


Is PVL a virulence factor in CA-MRSA pneumonia?

Mouse model

INFECTION:

- Balb/c 7-9 weeks old
- Intranasal inoculation using $2-6 \times 10^7$ CFU in solution



Mouse model

We look for:

Survival

Overall appearance

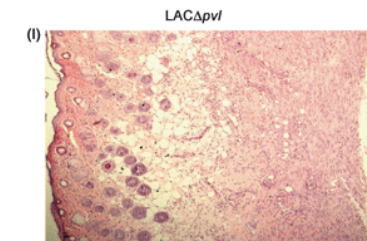
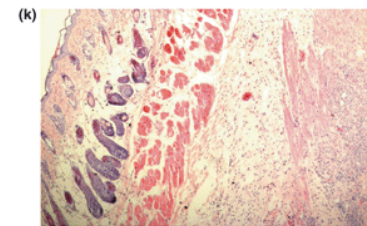
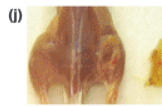
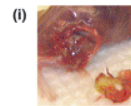
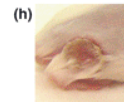
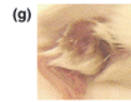
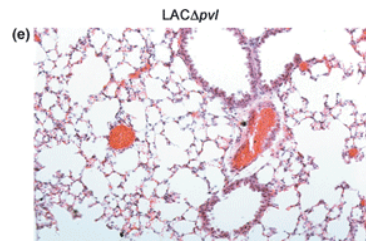
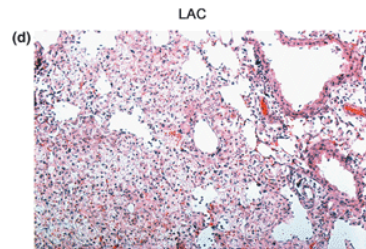
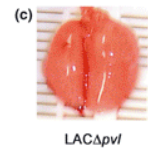
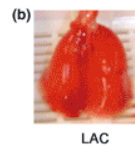
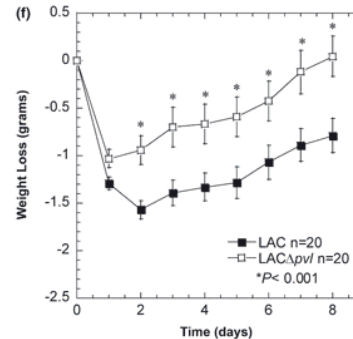
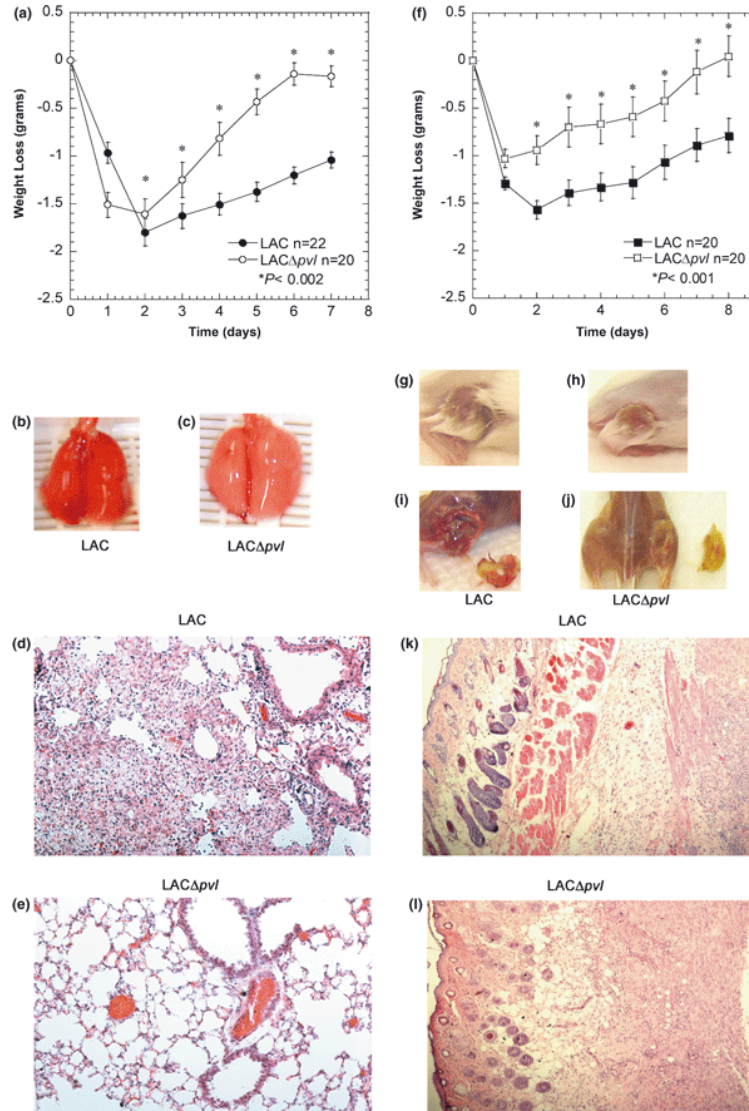
Weight loss

Bacteremia

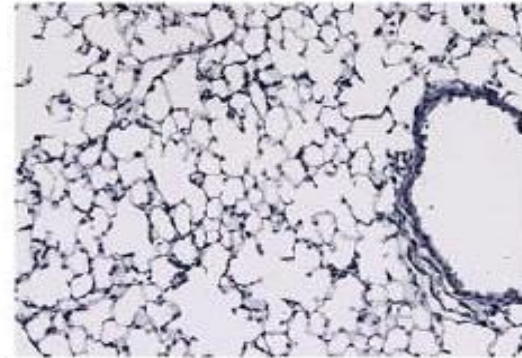
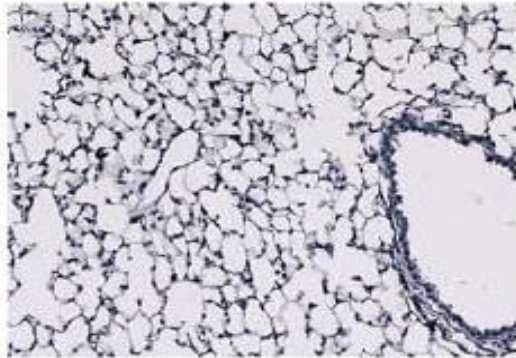
Gross lung morphology

Lung histology

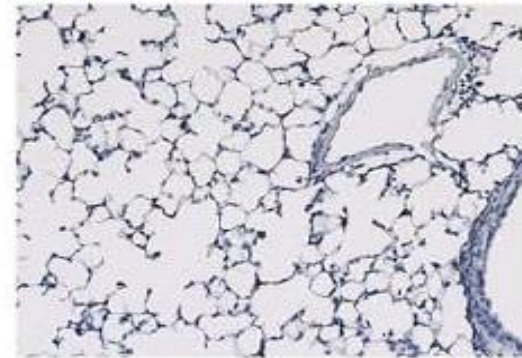
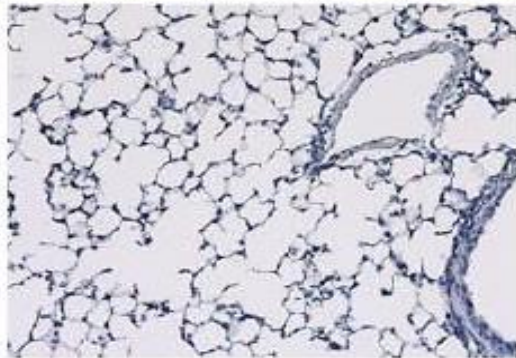
Staphylococcal clinical isolates can cause murine pneumonia



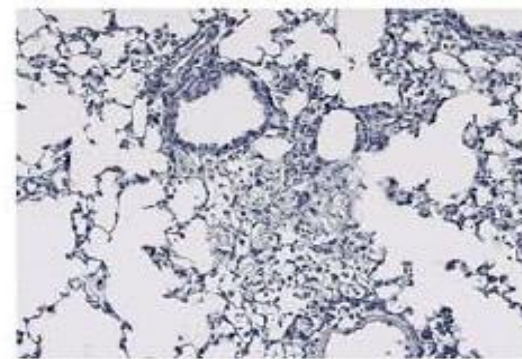
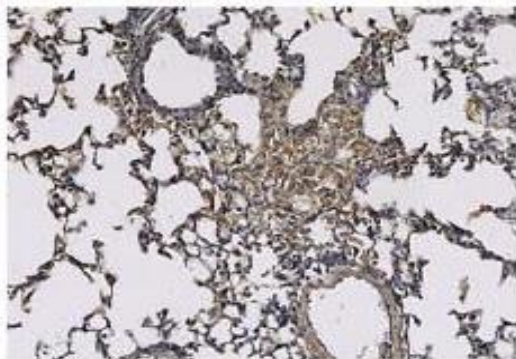
PVL is expressed *in vivo*



Parental

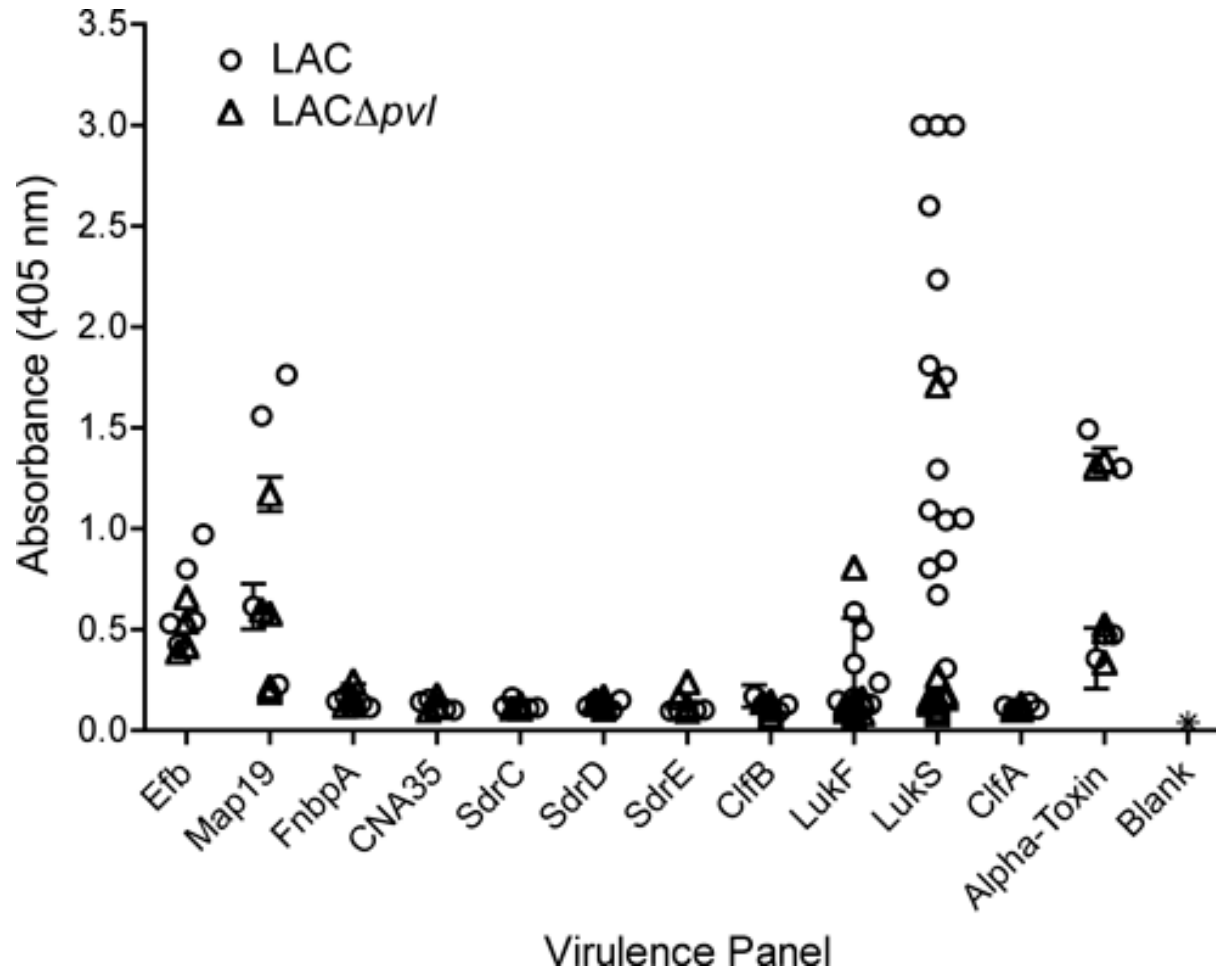


ΔPVL Phage

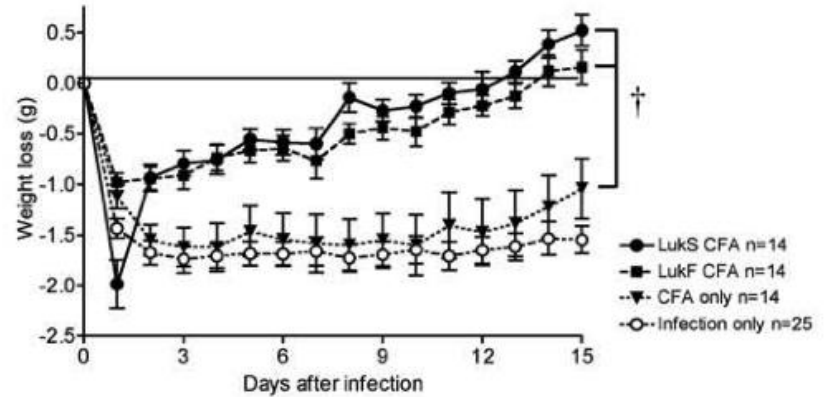
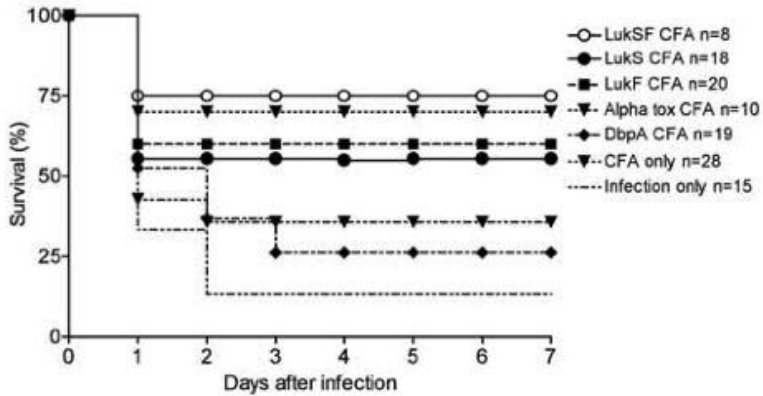


PVL Phage

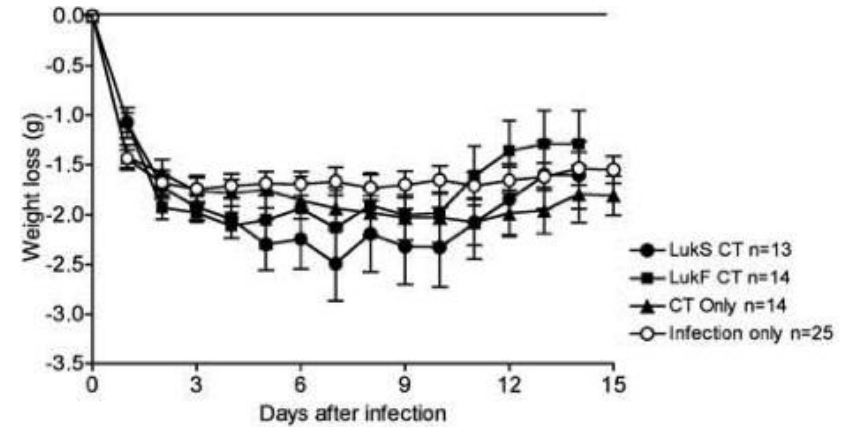
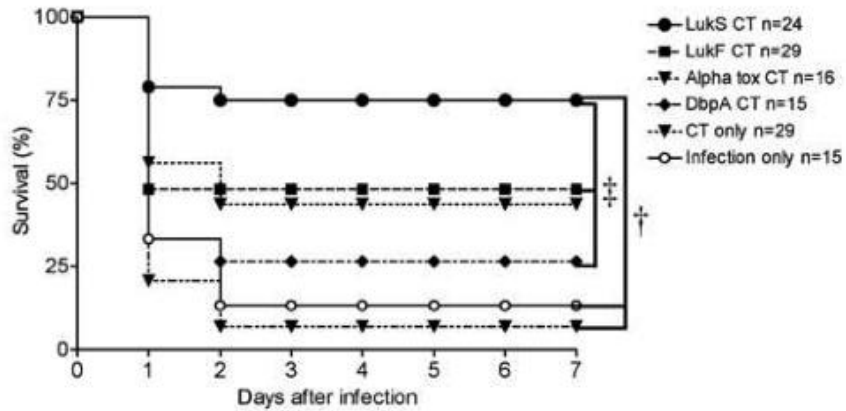
Antibody Response to *S. aureus* Virulence Factors Following Infection with USA300



I.D. Vaccination

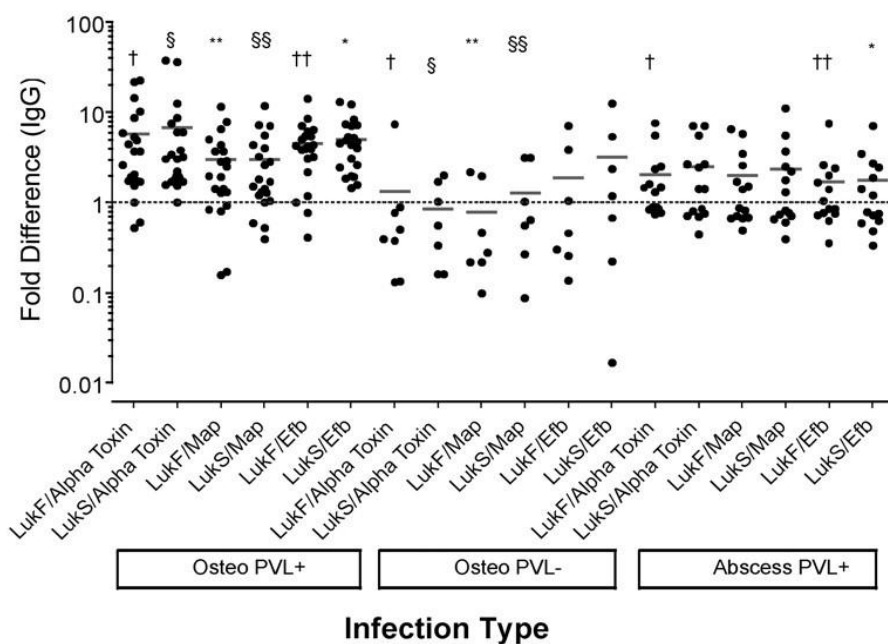
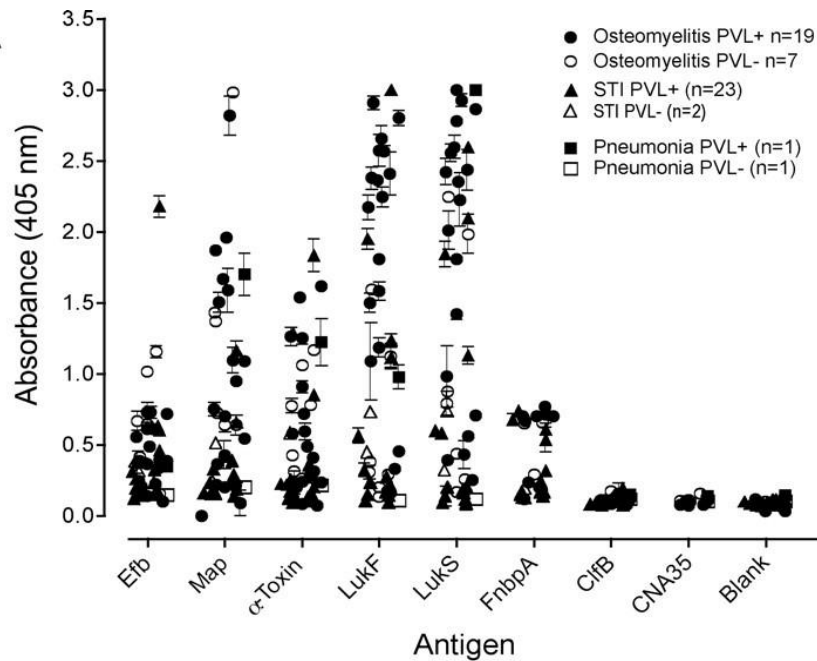


I.N. Vaccination

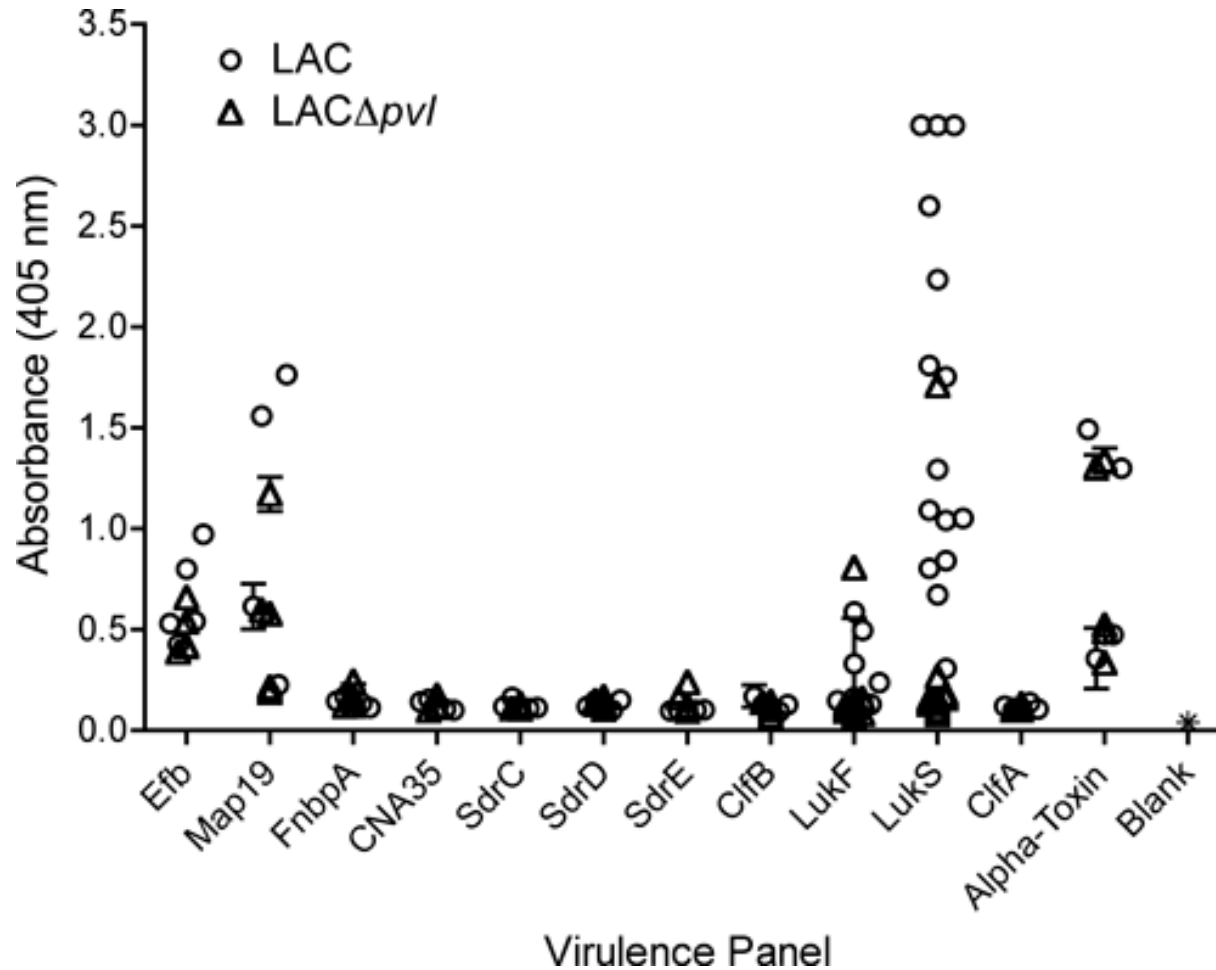


Route & Dose

Antibody Response to *S. aureus* Virulence Factors in Humans Following Infection with USA300



Antibody Response to *S. aureus* Virulence Factors Following Infection with USA300



PVL-positive isolates (identified by PCR)

- Skin abscesses

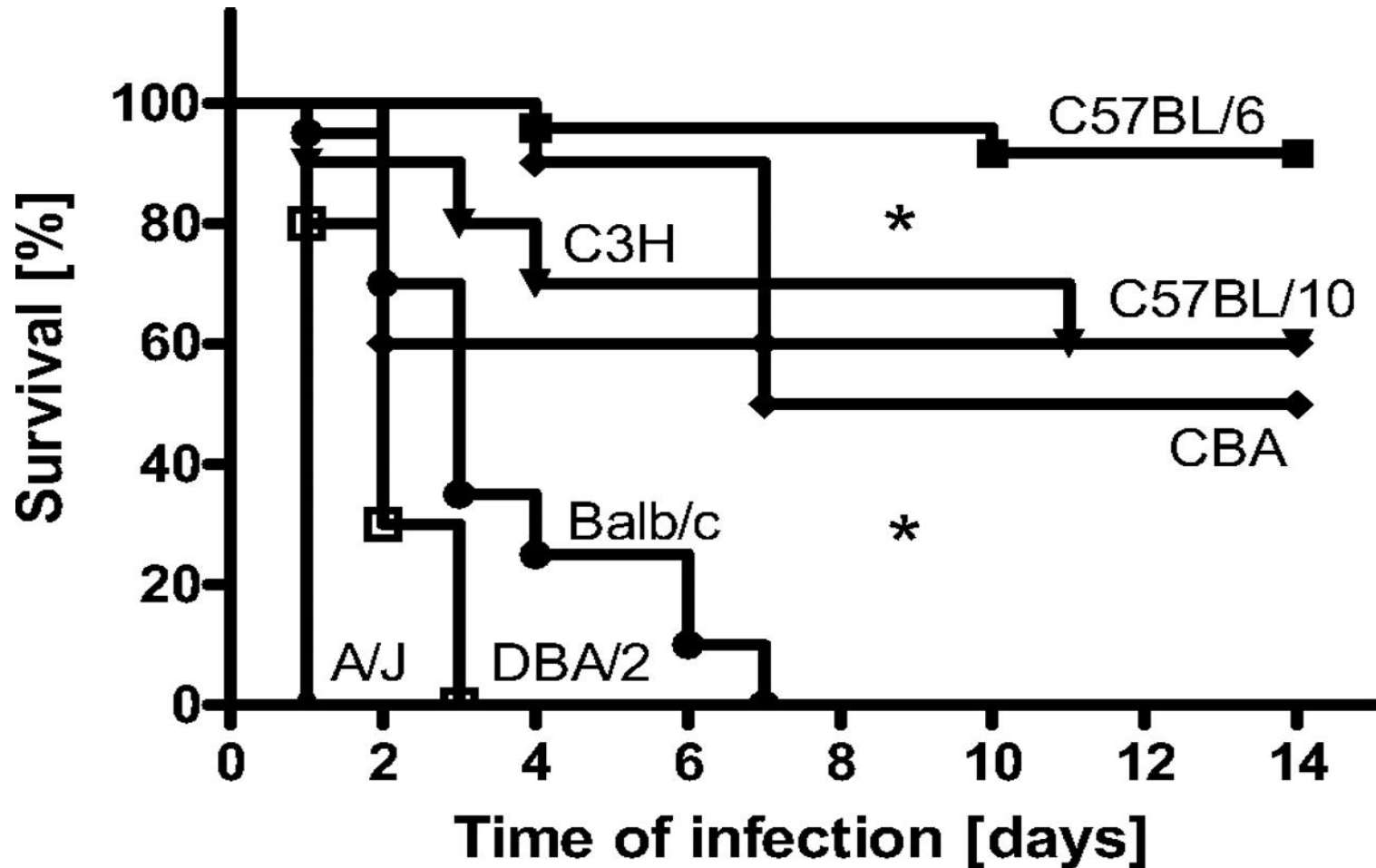
| Sampling | PVL gene | LukS-PV ($\mu\text{g/ml}$) | <i>mecA</i> |
|----------|-----------------|------------------------------|-------------|
| pus | Positive | >1 | 0 |
| pus | Positive | 0.4 | 0 |
| pus | Positive | >1 | 1 |
| pus | Positive | >1 | 0 |
| pus | Positive | 1.2 | 0 |
| pus | Positive | 1 | 0 |
| pus | Positive | 1.3 | 0 |
| pus | Positive | 1.5 | 1 |

PVL-negative isolates (identified by PCR)

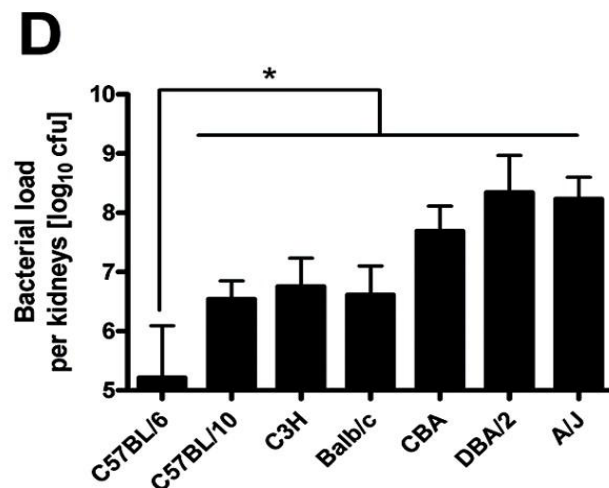
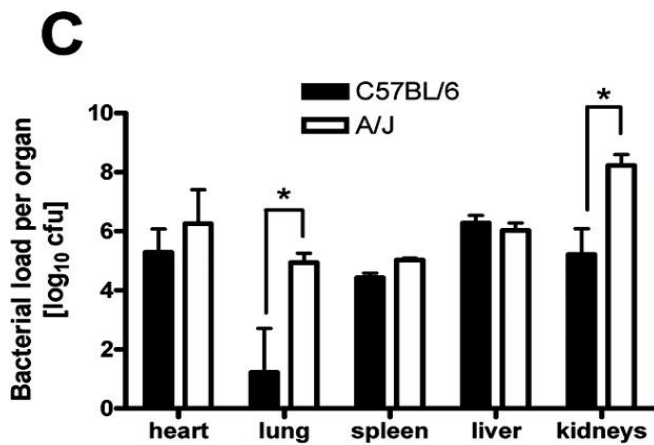
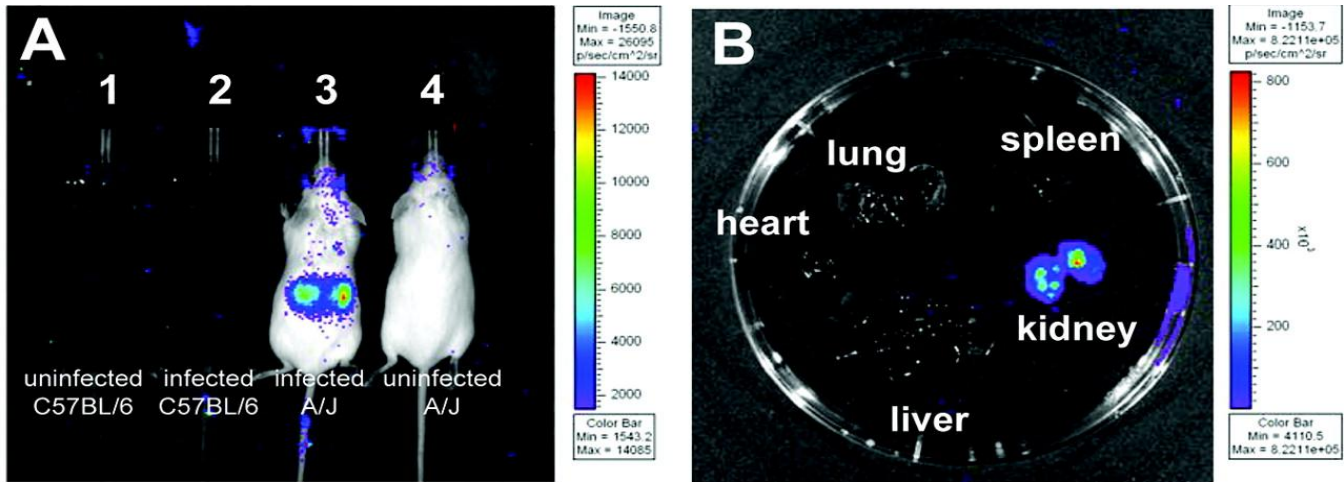
- **Pneumonia**
 - 16 BAL
 - 4 sputum
- **Skin abscesses**
 - 7 pus
- **LukS-PV was undetectable by ELISA**

Human Genetics and
S. aureus
carriage/disease

% Survival (4×10^7 cfu, i.v.)

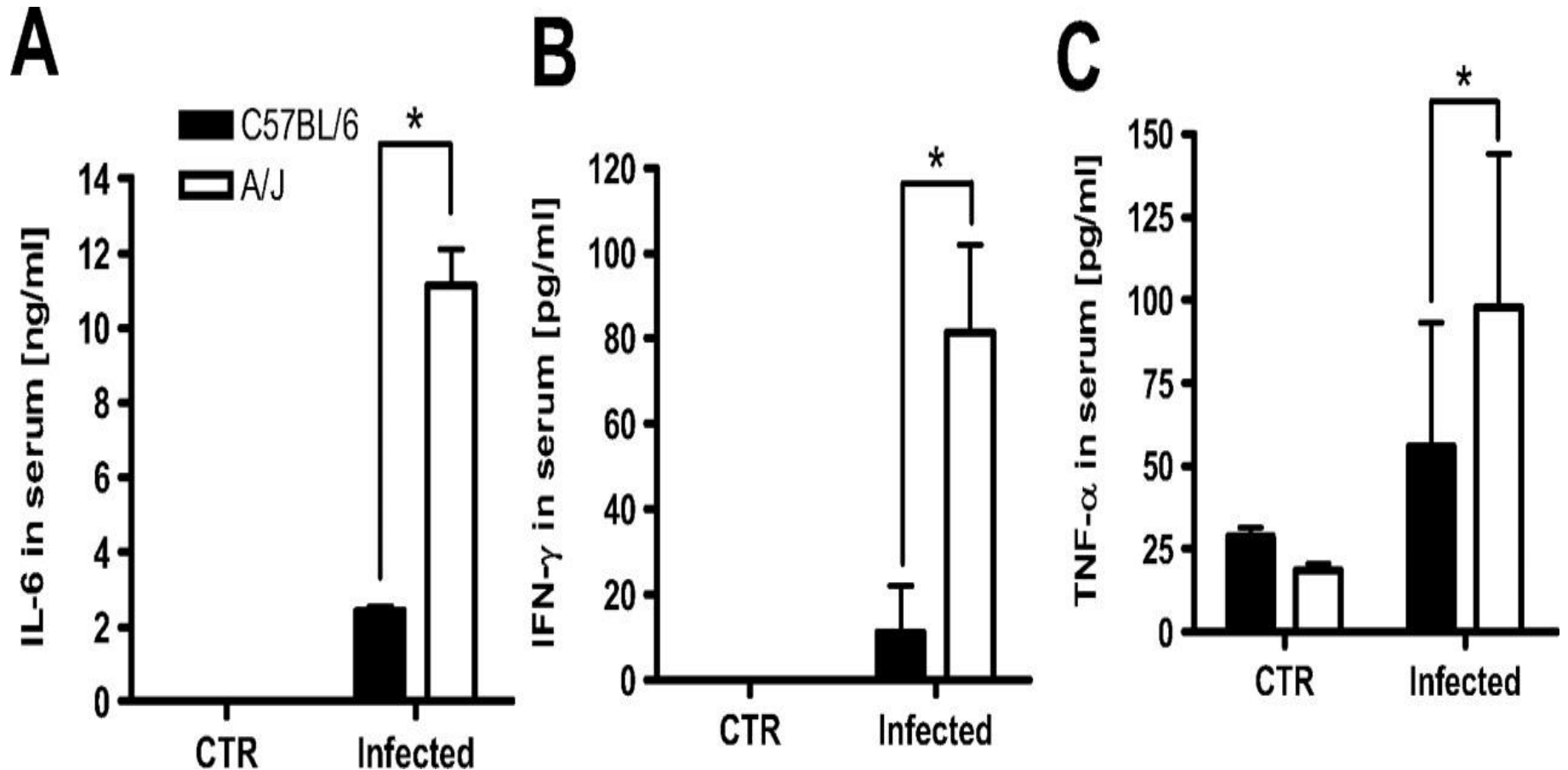


Bacterial Loads

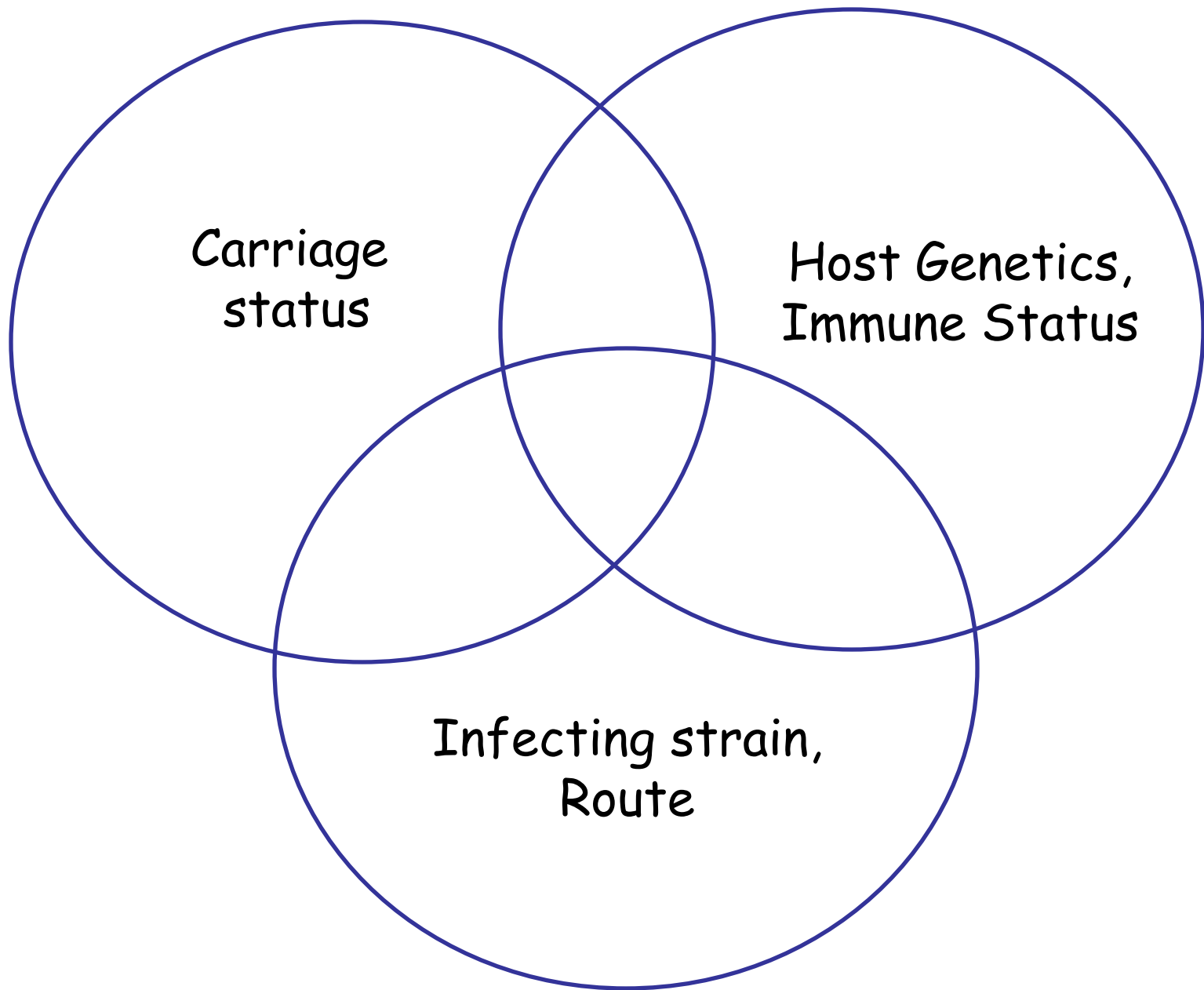


von kockritz-Blickwede, Am. J. Pathol., 2008

Cytokine Levels Post Infection



von kockritz-Blickwede, Am. J. Pathol., 2008



Carriage
status

Host Genetics,
Immune Status

Infecting strain,
Route

Host Polymorphisms in IL-4,
Complement Factor H, and C-
Reactive Protein: Association
with *S. aureus* carriage.

Methods

- Prevalence of persistent nasal carriage of *S. aureus* in 3851 participants aged 61-101.
 - Carriers, 18%
 - Non-carriers, 73%

Results

- Association between the -524 promoter polymorphism in IL-4 and nasal carriage of *S. aureus*.

- IL-4 (-524)

- C- 366 (91%)
- T- 38 (9%)

Future Directions

- Genome-Wide analysis of 4000 subjects defined by their *S. aureus* carriage.
 - Starr County
 - Pilot study: defined carriage for 84 patients

Inflammatory pathway (*i.e.*, cytokines, chemokines, complement, Toll receptors), matrix proteins (*e.g.*, fibronectin, collagen, fibrinogen).

S. aureus Vaccines

Target Groups for a *S. aureus* Vaccine

- Kidney patients who are receiving peritoneal dialysis surgical patients, particularly those undergoing lengthy cardiac and orthopedic procedures
- Trauma and burn patients
- Individuals undergoing invasive outpatient procedures
- Patients receiving an implanted medical device or prosthetic
- Newborns
- Individuals in long-term care
- Type 1 diabetics
- Immune-compromised individuals, such as cancer patients and patients with acquired immunodeficiency syndrome.

Don't give bacteria a free ride.



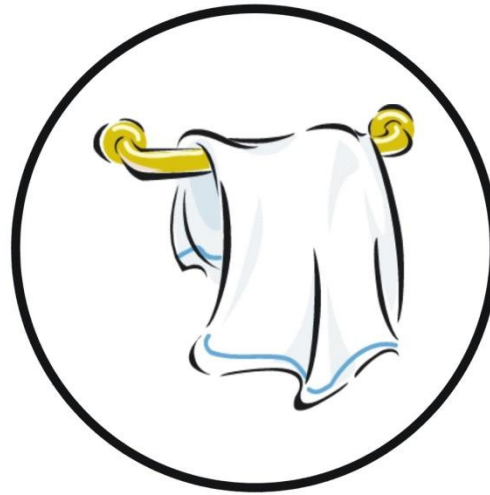
WASHING YOUR HANDS
WITH SOAP AND WATER
IS ONE OF THE BEST WAYS
TO PREVENT DISEASES.



www.cdc.gov/mrsa



Sharing isn't always caring.



SHARING PERSONAL ITEMS LIKE
TOWELS, RAZORS, OR TWEEZERS
CAN SPREAD DISEASES.



www.cdc.gov/mrsa



Laboratory Diagnosis

- Depends on type of infection
 - abscess
 - scrape the base of the abscess--abundance of bacteria
 - aspirated pus will have few organisms.
 - impetigo
 - bacteremia
 - few in the blood (< one bacteria/ml blood)
- *S. aureus* seen in the nasopharynx of patients with SSSS and in the vagina of patients with TSS.

Laboratory Diagnosis

- Clinical specimens should be inoculated onto nutritionally enriched agar media supplemented with sheep blood.
 - *S. aureus* ferments mannitol (other staphylococci do not)
 - *S. aureus* can grow in 7.5% NaCl (inhibits the growth of most other organisms)
- Biochemical tests
 - coagulase (BACTi™ Staph)
 - protein A
 - heat-stable nuclease
 - antibiogram patterns
- Other
 - PCR
 - phage typing

Treatment, Prevention, Control

- Antibiotics...
- Until recently, MRSA strains existed only in hospitals.
 - CA-MRSA
- Anti-MSCRAMM antibodies
- Handwashing
- Decolonization of hospital personnel.
 - mupirocin (Bactoban)

Treatment, Prevention, Control

- **Screening**
 - patients
 - hospital personnel
 - personnel with persistent carriage at sites other than the nose should be considered for referral for appropriate specialist management.
- **Skin Decolonization**
 - 4% chlorhexidine bodywash/shampoo
 - 7.5% povidone iodine
 - 2% triclosan
 - patients should bathe daily for 5 days with the chosen antiseptic detergent