

Introduction to Microbiology



Anas Abu-Humaidan
M.D. Ph.D.

Staphylococci

سورة الاحزاب

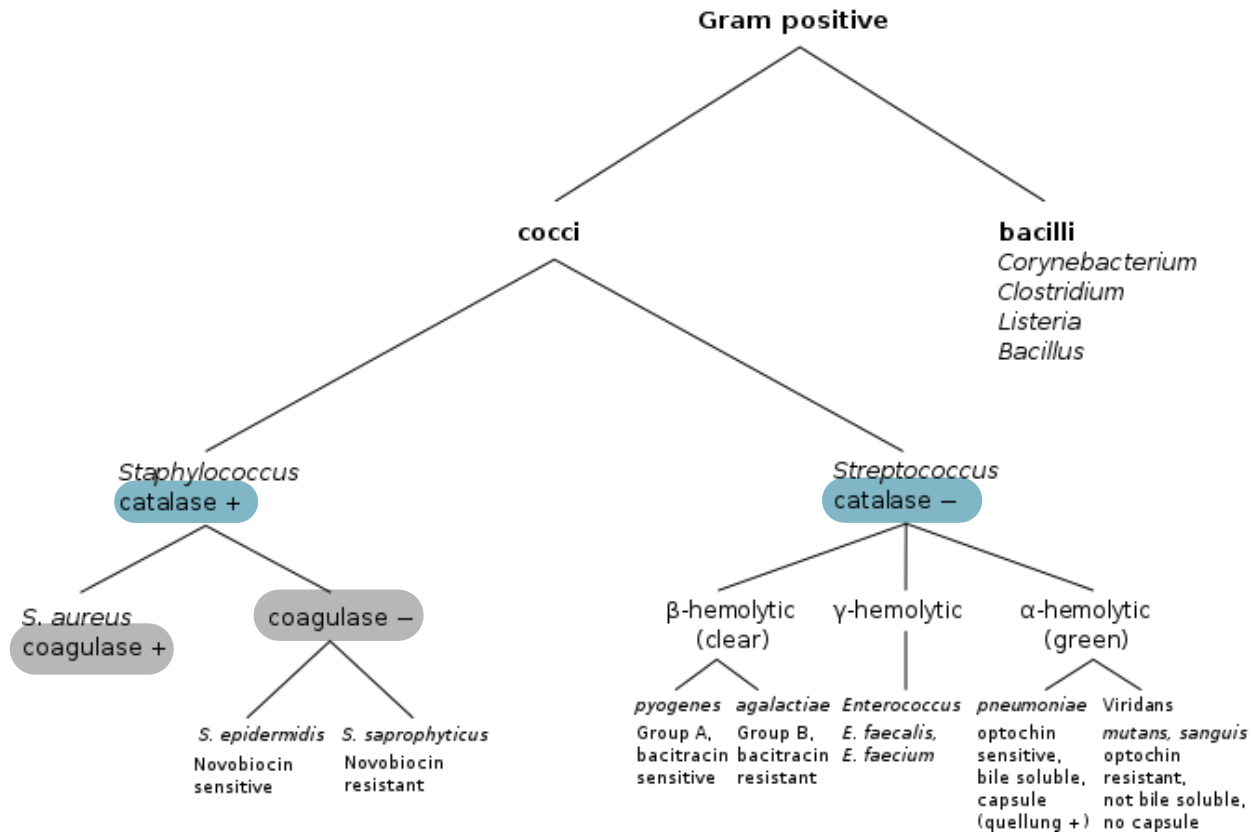
وَتَوَكَّلْ عَلَى اللَّهِ

وَكَفَى بِاللَّهِ وَكِيلًا

Overview

Bacterial genera that will be discussed this lecture are Gram positive cocci and that cause a variety of infections in the skin and mucus membranes, and can secrete a variety of toxins:

Staphylococci





- Gram +
- non-motile "lack the ability to move"
- Catalase + (to distinguish them from streptococci that are catalase-)

S. Aureus

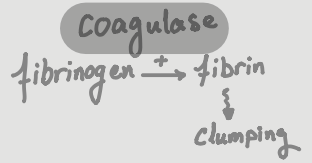
S. Epidermidis


S. Saprophyticus


Coagulase +


Coagulase -


Coagulase -



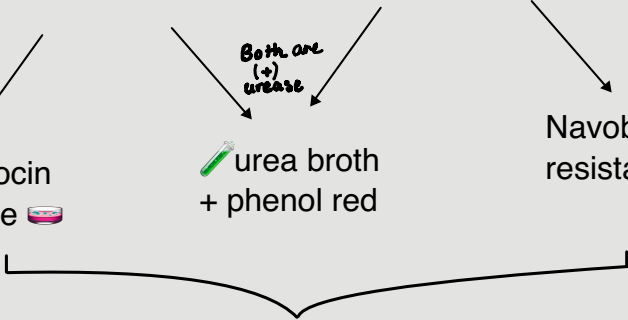
 mannitol salt agar
Golden yellow colonies

Navobiocin sensitive 

 urea broth
+ phenol red

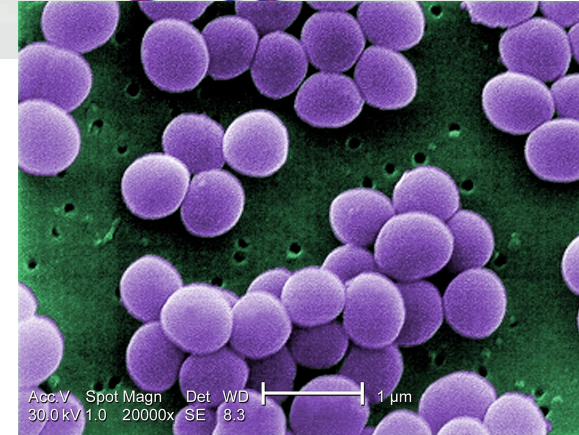
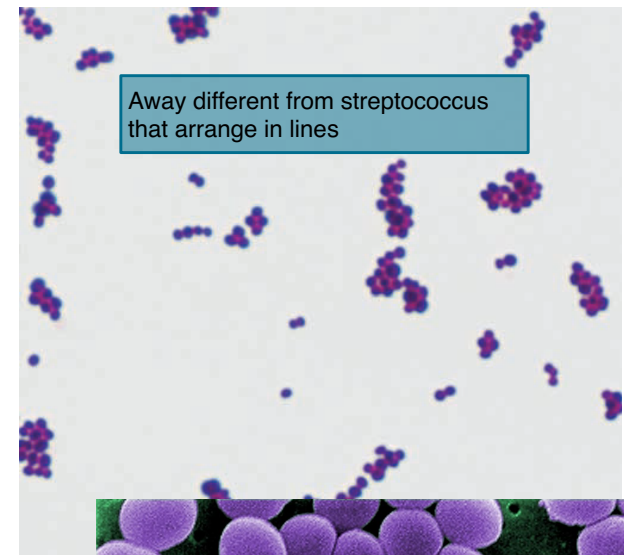
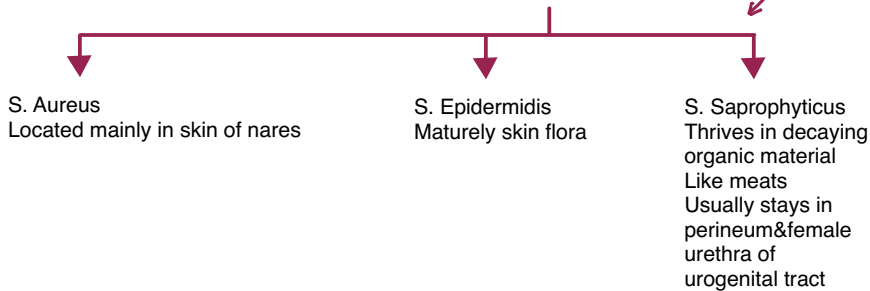
Navobiocin resistant 

Extra



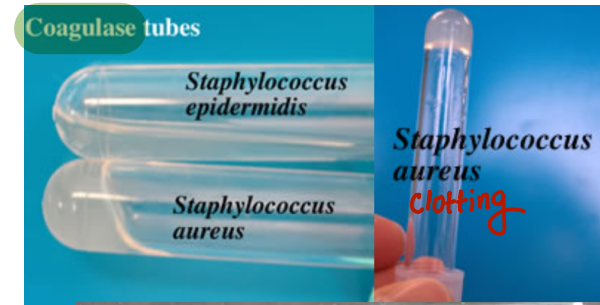
Staphylococci

- The staphylococci are **gram-positive spherical cells**, about 1 μm in diameter usually arranged in grapelike **irregular clusters**, it is **non-motile**.
Also, They can survive in both oxygen and non-oxygen environments
- The four most frequently encountered species of clinical importance are *Staphylococcus aureus*, *Staphylococcus epidermidis*, *Staphylococcus lugdunensis*, and *Staphylococcus saprophyticus*. Extra ↷



Staphylococci

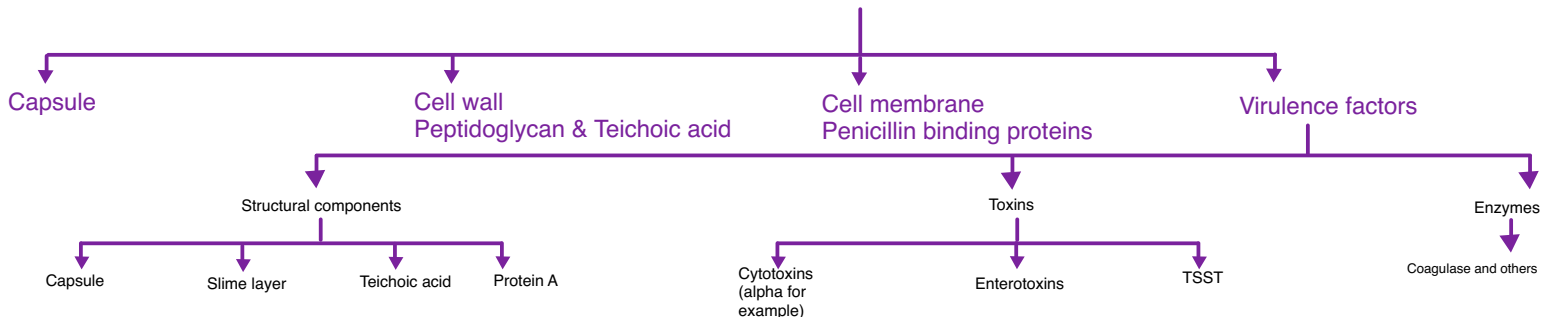
- *S aureus* is **coagulase positive**, The **(coagulase-negative staphylococci)** are {normal human microbiota. }
 - epidermidis
 - saprophyticus
- Staphylococci produce **catalase**, which **converts hydrogen peroxide into water and oxygen**. The catalase test differentiates the **staphylococci**, which are **positive**, from the **streptococci**, which are **negative**.
- *S aureus* usually **forms gray to deep golden yellow colonies**. *S epidermidis* colonies usually are **gray to white** on primary isolation
- **Various degrees of hemolysis** are produced by



Staphylococci / Structure and physiology

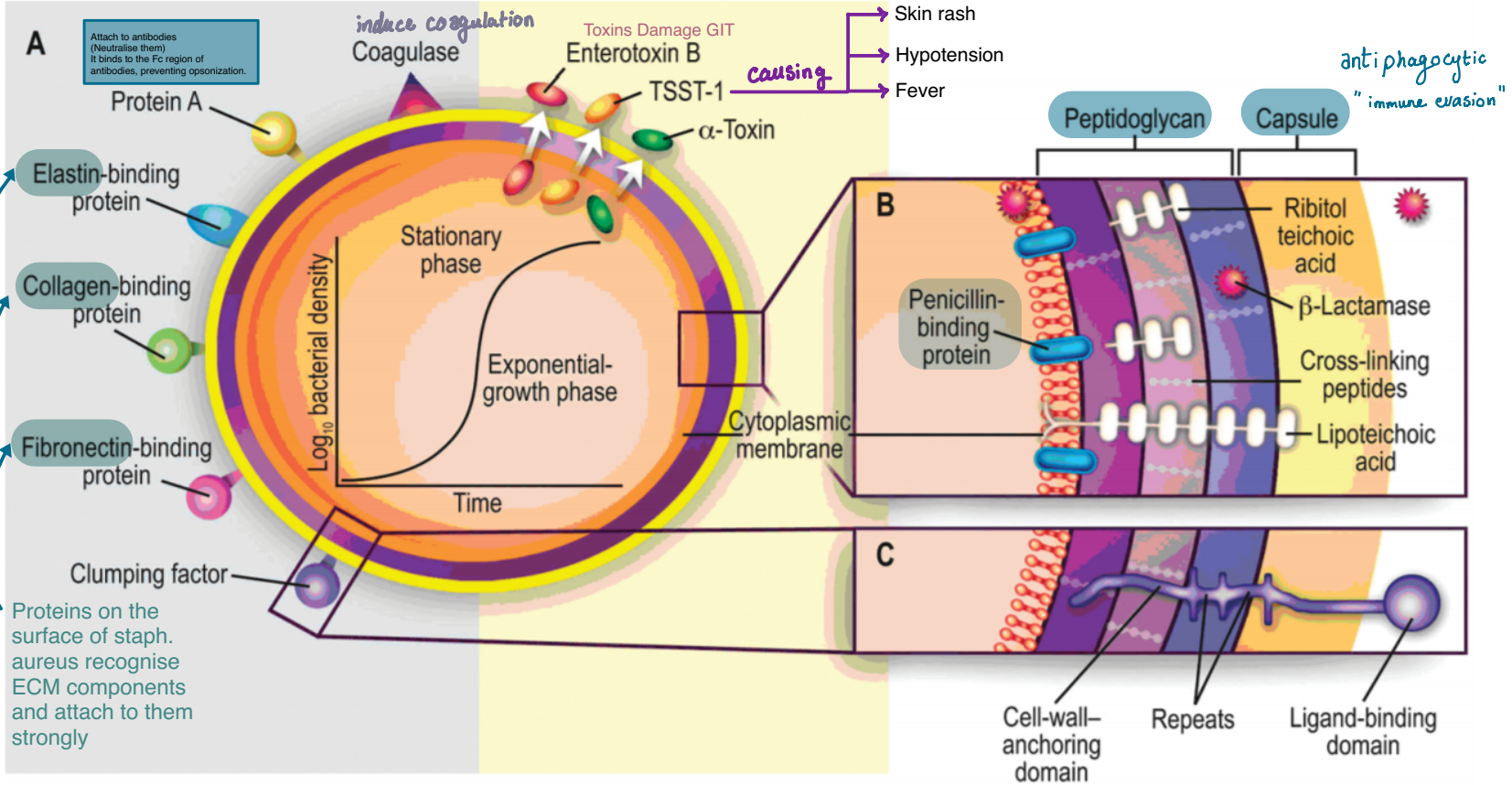
How they can cause damage?

- **Peptidoglycan in the cell wall** activate the immune response (it can be a chemoattractant for polymorphonuclear leukocytes, have endotoxin-like activity, and activate complement.)
Neutrophils
- Bacterial attachment to host cells is mediated by **MSCRAMM (microbial surface components recognizing adhesive matrix molecules) proteins**. and these are important **virulence factors**. (e.g. Protein A, clumping factor)
- **Teichoic acids** are cross-linked to the peptidoglycan and can be antigenic.
- **Clumping factor A** is a fibrinogen-binding protein present on the surface of *S. aureus* that binds to fibrinogen and coats the surface of the bacterial cells with fibrinogen molecules, additionally complicating the recognition process.



Surface proteins (exponential-growth phase)

Secreted proteins (stationary phase)



Not all staph. Aureus have the same components !!!
Some are more virulent than others , depending on how much virulence factors they have

Staphylococci / Epidemiology

Distribution in community

Staphylococci, particularly *S epidermidis*, are members of the normal microbiota of the human skin and respiratory and gastrointestinal tracts.

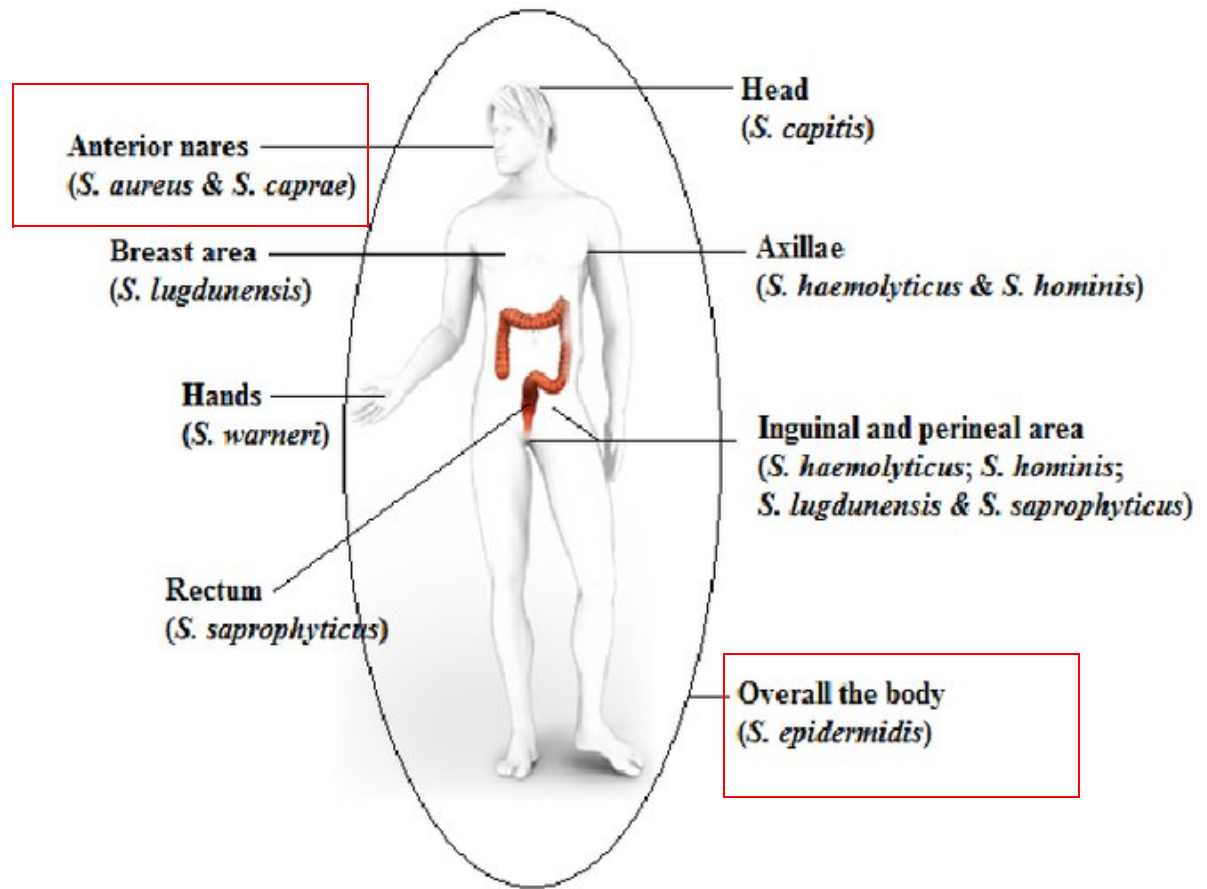
Nasal carriage of *S aureus* occurs in 20–50% of humans, with a **higher incidence** reported for hospitalized patients, medical personnel, persons with eczematous skin diseases. Staphylococci are also found regularly on clothing, bed linens, and other **fomites** in human environments.

ubiquitous

It somehow destroys the skin barrier, this enhances the chance of aureus to colonise

Aureus can be found as a coloniser without causing disease

Epidermidis is found in all humans
Aureus found in some



The epidemiology and molecular characterization of methicillin-resistant staphylococci sampled from a healthy Jordanian population

A G Al-Bakri 1, H Al-Hadithi, V Kasabri, G Othman, A Kriegeskorte, K Becker

Abstract

The prevalence of natural carriage and molecular epidemiology of methicillin-resistant *Staphylococcus aureus* (MRSA) and methicillin-resistant coagulase-negative staphylococci (MR-CoNS) isolates in a Jordanian community were investigated. The MRSA nasal carriage rate in 227 healthy volunteers was 7.5% and the majority (81%) of MRSA harboured the resistance element SCCmec type IVe and were of a novel spa type t9519 (76%); other significant spa gene types were t223 (14.7%) and t044 (5.9%).

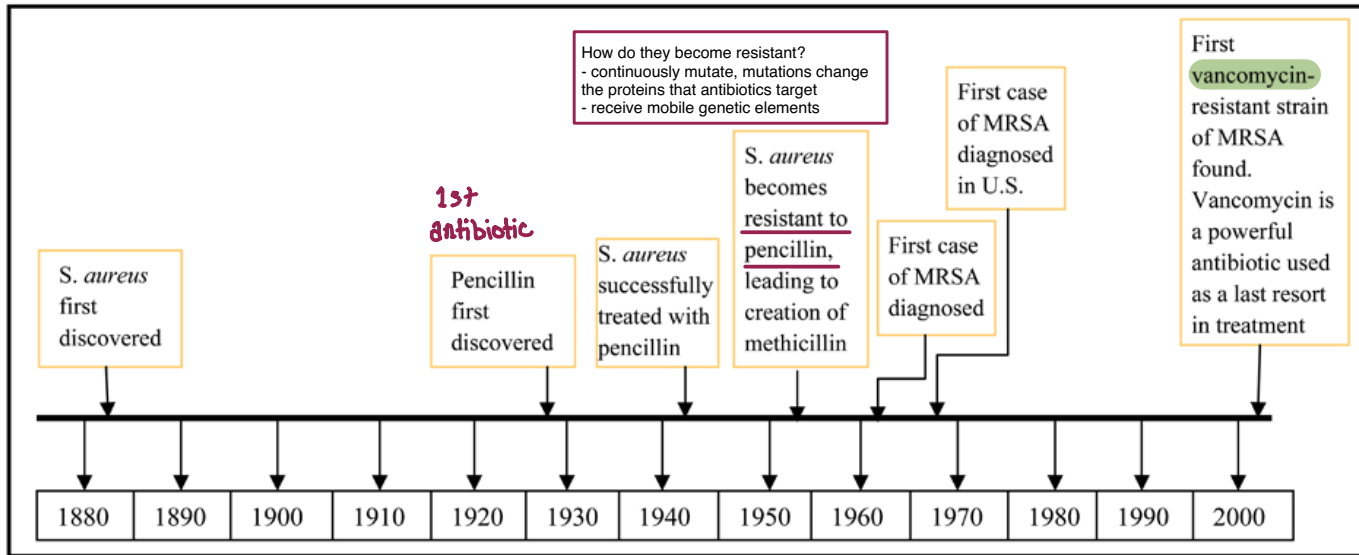
All MRSA isolates were susceptible to other classes of antibiotics, and tested positive for at least three virulence factor encoding genes, but only two harboured the pvl gene. MR-CoNS carriage was 54.2% and these isolates were characterized by single, double and untypable SCCmec elements, with *Staphylococcus epidermidis* SCCmec type IVa predominating. Of eight subjects with nasal co-colonization of MR-CoNS + MRSA, three shared SCCmec type IV in both groups of organisms. This is the first report of methicillin-resistant staphylococci carriage in a Jordanian community and its findings are important for epidemiological study and infection control measures of these organisms.

mobile genetic element found in *Staphylococcus* bacteria. This means it's a segment of DNA that can move between bacterial cells, allowing it to spread antibiotic resistance genes. The SCCmec element carries the *mecA* gene, which provides resistance to methicillin antibiotics

↪ a gene that carries resistance to methicillin antibiotic

SCCmec, or **staphylococcal cassette chromosome *mec***, is a mobile genetic element of *Staphylococcus* bacterial species. This genetic sequence includes the *mecA* gene coding for resistance to the antibiotic methicillin and is the only known way for *Staphylococcus* strains to spread the gene in the wild by horizontal gene transfer.

MRSA is associated with hospitals, environments
And associated with more invasive and serious infections
of *s.aureus* that can reach blood causing bacteremia



Beginning in the 1980s, strains of Methicillin-resistant *Staphylococcus aureus* (MRSA) spread rapidly in susceptible hospitalized patients, dramatically changing the therapy available for preventing and treating staphylococcal infections.

MRSA began as a hospital-acquired infection, but has become community-acquired as well as livestock-acquired.

People with compromised immune systems (elderly, diabetics, HIV/AIDS), hospitalized patients and children are some of the susceptible groups to MRSA.

Staphylococci / Clinical correlations

A skin erythematous infection refers to a skin infection characterized by erythema

A localized staphylococcal infection appears as a “**pimple,**” **hair follicle infection,** or **abscess.** There is usually an intense, localized, painful inflammatory reaction that undergoes central suppuration and heals quickly when the pus is drained.



Aggregation of PMNS and macrophages
They come to the site of inflammation
skin infection characterized by **erythema**



→ infects the epidermis only

Impetigo: localized cutaneous infection characterized by pus-filled vesicle on an erythematous base

Folliculitis: impetigo involving hair follicles

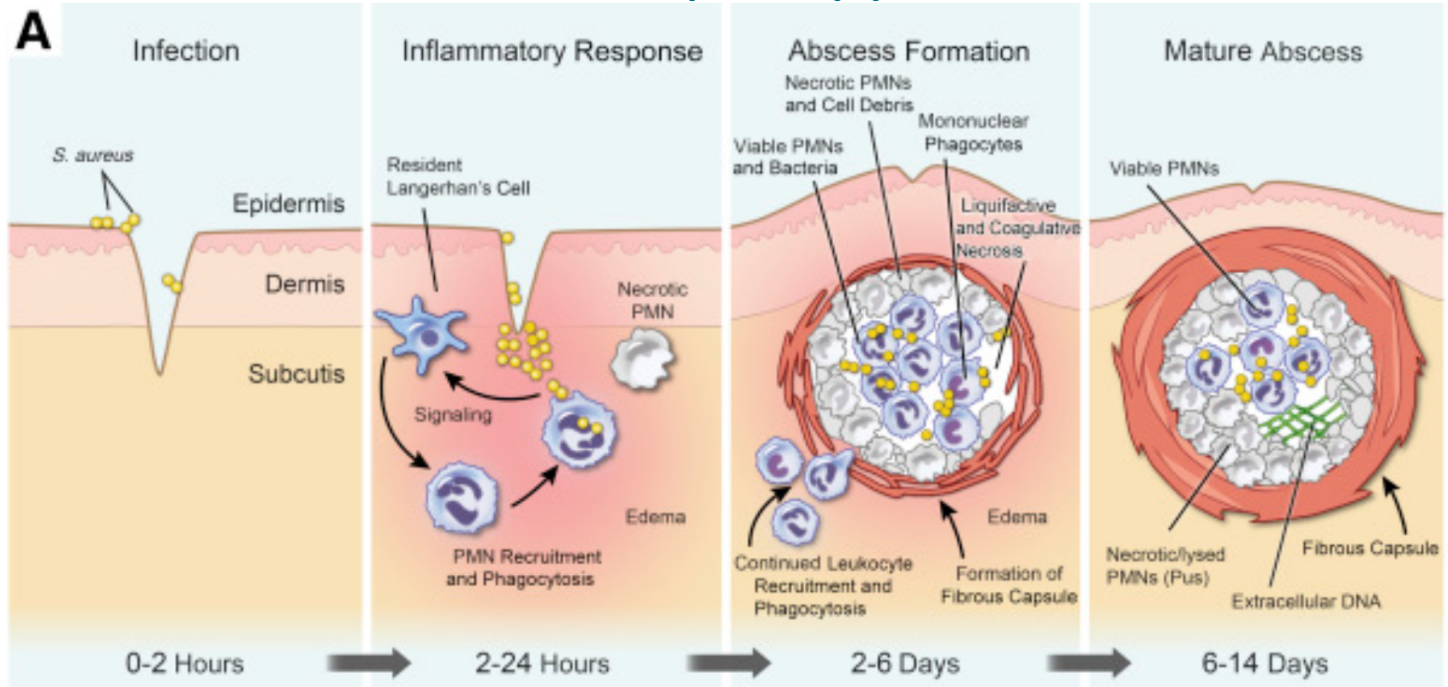
Furuncles or boils: large, painful, pus-filled cutaneous nodules

Carbuncles: Coalescence of furuncles with extension into subcutaneous tissues and evidence of systemic disease (fever, chills, bacteremia)


→ many furuncles together



When abscess forms it's harder for antibiotics to act
No BV feed this part
Surgical removal && giving antibiotics in these cases



Staphylococci / Clinical correlations / S. aureus bacteremia

Bacteria reach the blood 

Simultaneously, SA is a leading cause of **bacteremia** and **infective endocarditis** (IE) as well as **osteoarticular**, skin and soft tissue, **pleuropulmonary**, and device-related infections.

↻ before sepsis
↻ S. aureus reach the heart
reaching bones & joints
in hospitalized patients, S. aureus cause infections of respiratory tract

Clinical infections with S. aureus will likely remain both common and serious. Not only have there been waves of increasing antimicrobial resistance (827), but the spectrum of clinical disease also continues to change. In the past 2 decades, we have witnessed two clear shifts in the epidemiology of S. aureus infections: first, ¹ a growing number of health care-associated infections, particularly seen in IE and prosthetic device infections, and second, an ² epidemic of community-associated SSTIs driven by strains with particular virulence factors. There is no doubt that there will continue to be a shifting landscape in the interactions between host and pathogen in the decades to come.

How many individuals their s.aureus in blood were MRSA

Region (reference)	% of MRSA cases in cohort	% of HCA cases in cohort	No. (%) of cases with focus of infection							Total no. of cases
			Infective endocarditis	Osteoarticular	SSTI	Pleuropulmonary	Line related	No focus/unknown	Other	
Central Australia (32)	21.6	25.6	9 (7.2)	20 (16)	42 (34)	11 (8.8)	9 (7.2)	30 (24)	4 (3.2)	125
Australia (59)	24.8	79.1	433 (6)	956 (13)	1,415 (20)	519 (7.2)	1,387 (19)	1,100 (15)	1,421 (20)	7,231
Sydney, Australia (65)	100	92	15 (3.8)	37 (9.3)	80 (20)	52 (13)	140 (35)	40 (10)	35 (8.8)	399
Calgary, Canada (12) ^b	11.3	75.3	79 (5.5)	227 (16)	224 (16)	220 (15)		586 (41)	104 (7.2)	1,440
Missouri, USA (64)	100	92.6	0 (0)	0 (0)	39 (24)	0 (0)	37 (23)	70 (43)	17 (10)	163
New York, USA (61)	100	97.9	91 (14)	72 (11)	112 (17)	55 (8.4)	302 (46)	0 (0)	20 (3.1)	652
Birmingham, UK (66)	100	99.5	6 (3.1)	3 (1.5)	37 (19)	0 (0)	73 (37)	68 (35)	8 (4.1)	195
Italy (57)	53.9	85.5	0 (0)	0 (0)	14 (9.3)	7 (4.6)	23 (15)	104 (69)	3 (2)	151
Israel (56)	42.8	100	55 (4.4)	71 (5.6)	294 (23)	144 (11)	172 (14)	298 (24)	227 (18)	1,261
Thailand (58)	27.6	55.1	8 (11)	9 (12)	20 (27)	16 (22)	10 (14)	0 (0)	10 (14)	73
South Korea (63)	100	95.1	9 (3.4)	16 (6)	35 (13)	24 (9)	132 (49)	36 (13)	16 (6)	268
Japan (62)	100	NA	0 (0)	0 (0)	17 (15)	10 (8.7)	27 (23)	23 (20)	38 (33)	115
Multisite (60)	11.7	NA	282 (8.3)	456 (13)	502 (15)	178 (5.2)	942 (28)	641 (19)	394 (12)	3,395
Total										15,468

^aThe mean percentages of patients for each primary focus of infection from all the studies were as follows: 5% for infective endocarditis, 8% for osteoarticular, 19% for SSTI, 9% for pleuropulmonary, 26% for line related, 24% for no focus/unknown, and 11% for other foci. MRSA, methicillin-resistant *S. aureus*; HCA, health care associated; SSTI, skin and soft tissue infection.

^bLine-related bacteremia was not reported in this study.

Staphylococci / Clinical correlations / Toxin mediated

Staphylococcal food poisoning, one of the most common foodborne illnesses, is an **intoxication** rather than an infection. Disease is caused by **heat stable bacterial toxin** present in food rather than from a direct effect of the organisms on the patient. With a **short incubation period** (1–8 hours); violent nausea, vomiting, and diarrhea; and **rapid convalescence**.

Between entry and onset of symptoms

recovery stage

Staphylococcal scalded skin syndrome is a condition which predominantly affects **infants** and **children** and causes a spectrum of skin lesions.

Toxin-Mediated Diseases

Scalded skin syndrome: Disseminated desquamation of epithelium in infants; blisters with no organisms or leukocytes

Food poisoning: After consumption of food contaminated with heat-stable (enterotoxin) rapid onset of severe vomiting, diarrhea, and abdominal cramping, with resolution within 24 hours

Toxic shock: multisystem intoxication characterized initially by fever, hypotension, and a diffuse, macular, erythematous rash; high mortality without prompt antibiotic therapy and elimination of the focus of infection

TSS one of superantigens, can directly kill immune cells or stimulate them in a high degree where they become inefficient



Staphylococci / Clinical correlations / Coagulase negative

- *S. epidermidis* infections are ^{why?} difficult to cure [?] because they occur in prosthetic devices where the bacteria can sequester themselves in a biofilm. staphylococci are a major cause of endocarditis of artificial valves.
- More than 50% of all infections of catheters and shunts are caused by coagulase-negative staphylococci. These infections have become a major medical problem because long-dwelling catheters and shunts are used commonly for the medical management of critically ill patients.



يعني فوق المرض مرض

The catheter should be removed and use a new one

Coagulase-Negative *Staphylococcus* Species

Wound infections: Characterized by erythema and pus at the site of a traumatic or surgical wound; infections with foreign bodies can be caused by *S. aureus* and coagulase-negative staphylococci

Urinary tract infections: Dysuria and pyuria in young sexually active women (*S. saprophyticus*), in patients with urinary catheters (other coagulase-negative staphylococci), or following seeding of the urinary tract by bacteremia (*S. aureus*)

Catheter and shunt infections: Chronic inflammatory response to bacteria coating a catheter or shunt (most commonly with coagulase-negative staphylococci)

Prosthetic device infections: Chronic infection of device characterized by localized pain and mechanical failure of the device (most commonly with coagulase-negative staphylococci)

Summary...

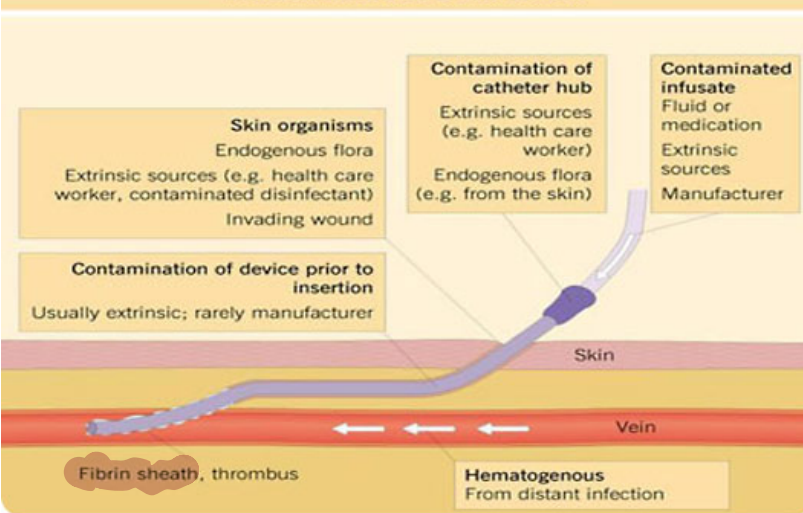
Some catheters could move some microbiota and s.Epidermidis from skin into deeper layers

→ s.aureus could also move but s.Epidermidis is the commonest

What should we do?

- disinfection (sterilisation) before inserting catheter
- hygiene methods should be applied by healthcare workers

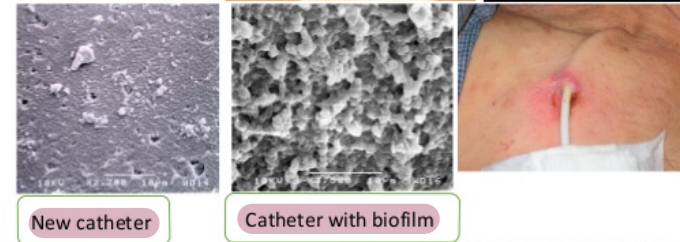
POTENTIAL ROUTES OF INFECTION



Catheter Exit Site infection



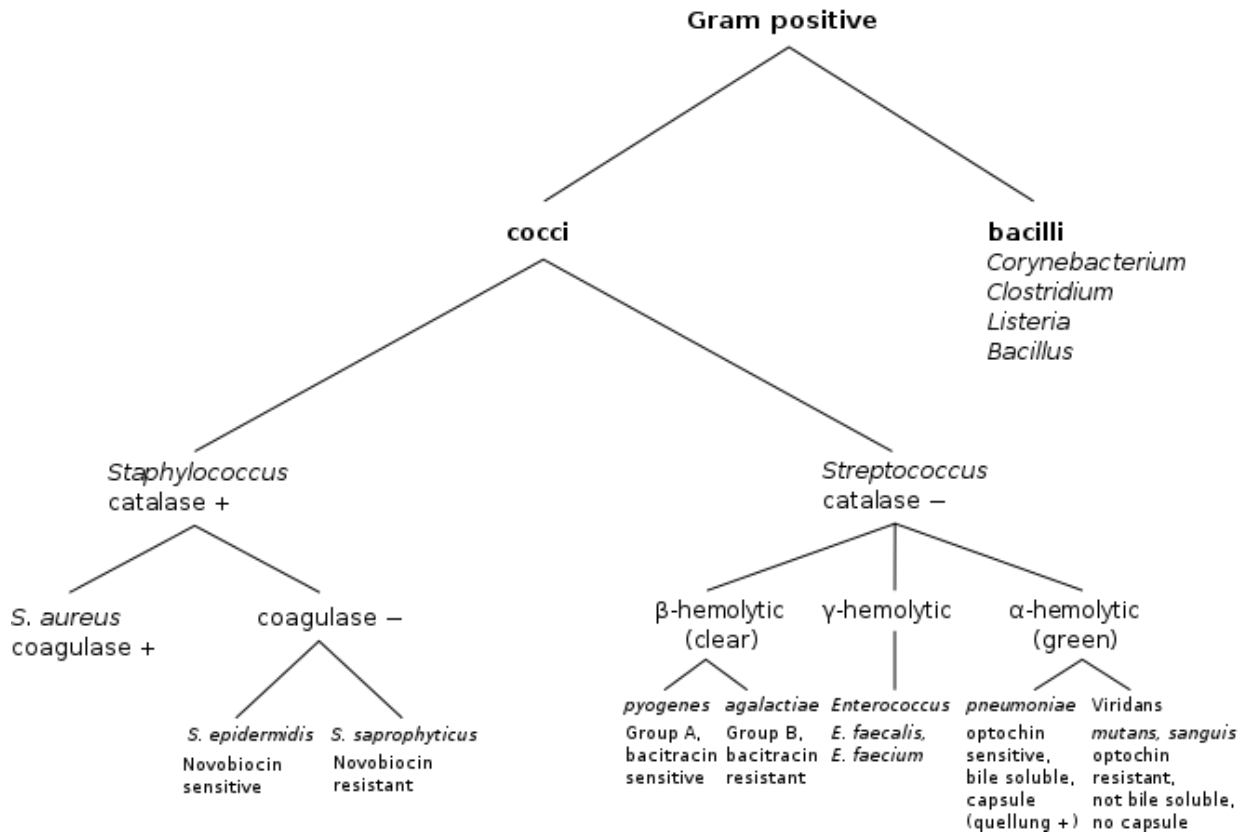
Catheter Tunnel infection



New catheter

Catheter with biofilm

Photo provided by Stephanie Booth, used with permission

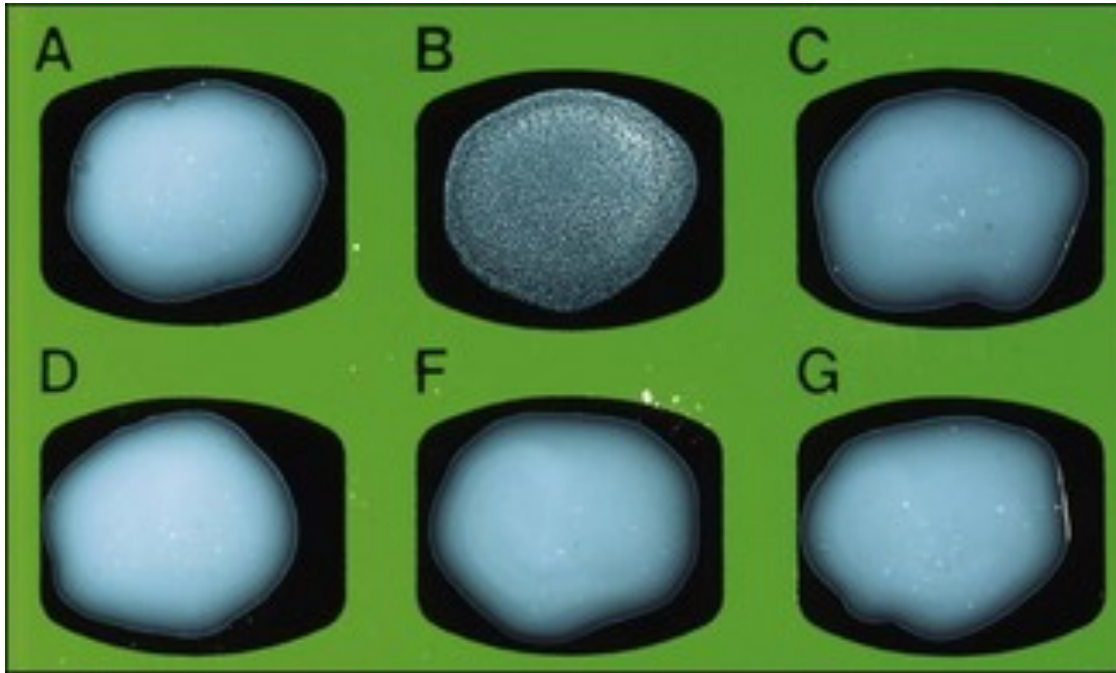


Streptococci / Classification

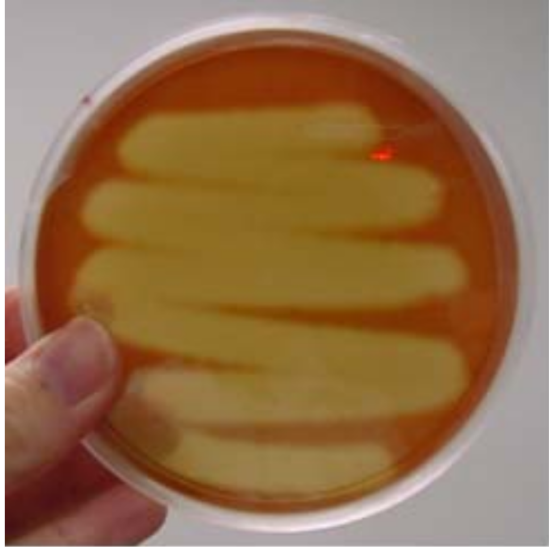
I think this will be recorded later

- The classification of more than 100 species within the genus *Streptococcus* is complicated because three different overlapping schemes are used:
 - (1) serologic properties: **Lancefield groupings** (originally A to W);
 - (2) **hemolytic patterns**: complete (beta [β]) hemolysis, incomplete (alpha [α]) hemolysis, and no (gamma [γ]) hemolysis;
 - (3) **biochemical (physiologic) properties.**
- The most important pathogenic streptococcal species for humans include *Streptococcus pyogenes* (group A streptococcus/ GAS) , *Streptococcus agalactiae* (GBS), ~~group D streptococcus (enterococci)~~, *Streptococcus pneumoniae*, and *Streptococcus viridans*.

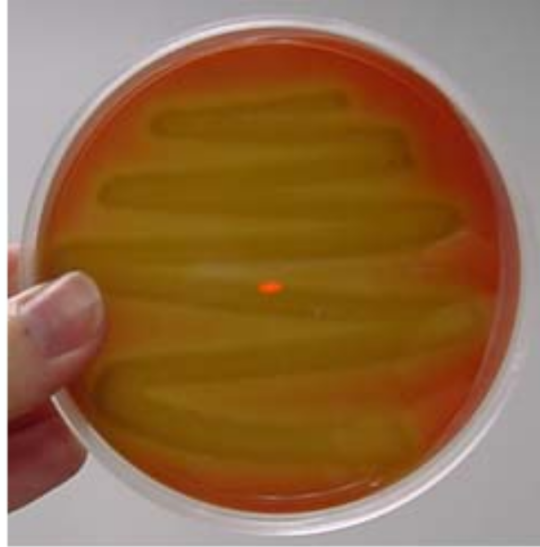
Streptococci / Classification / Lancefield groupings



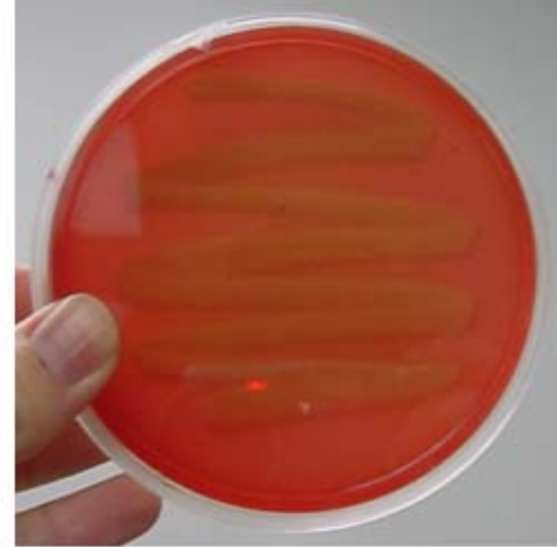
Streptococci / Classification / hemolytic patterns



Beta Hemolysis

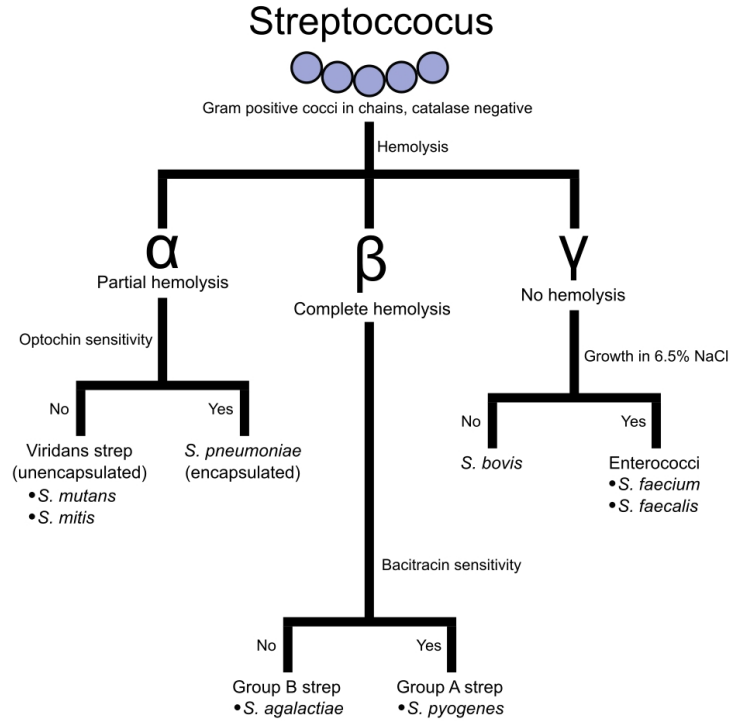


Alpha Hemolysis



Gamma Hemolysis

Streptococci / Classification / biochemical (physiologic) properties



© Lineage

Moises Dominguez

1. Which of the following *Staphylococcus* species is most commonly associated with human infections?

- A. *Staphylococcus epidermidis*
- B. *Staphylococcus saprophyticus*
- C. *Staphylococcus aureus*
- D. *Staphylococcus haemolyticus*

2. *Staphylococcus aureus* is known for its ability to produce several virulence factors. Which of the following toxins is primarily associated with toxic shock syndrome?

- A. Hemolysin
- B. Enterotoxin
- C. Exfoliative toxin
- D. Toxic shock syndrome toxin-1 (TSST-1)

3. Which test is commonly used to differentiate *Staphylococcus aureus* from other staphylococcal species?

- A. Catalase test
- B. Coagulase test
- C. Oxidase test
- D. Gram stain

4. *Staphylococcus epidermidis* is part of the normal skin flora but can be pathogenic under certain conditions. Which type of infection is it commonly associated with?

- A. Urinary tract infections
- B. Skin abscesses
- C. Device-associated infections
- D. Gastrointestinal infections

5. Which of the following statements about Methicillin-resistant *Staphylococcus aureus* (MRSA) is true?

- A. MRSA is only found in healthcare settings. it has spread to the community (community-associated MRSA) and can also be found in livestock (livestock-associated MRSA).
- B. MRSA produces beta-lactamase, which breaks down methicillin. MRSA's methicillin resistance is due to the mecA gene and the altered PBP2a protein, not the production of beta-lactamase.
- C. MRSA is resistant to all antibiotics. Not vancomycin 😊
- D. MRSA has an altered penicillin-binding protein (PBP2a) that reduces methicillin binding.

6. *Staphylococcus aureus* can cause food poisoning. Which of the following best describes the mechanism of food poisoning caused by *S. aureus*?

- A. Production of an endotoxin that contaminates food
- B. Production of a heat-stable enterotoxin in food
- C. Production of an exotoxin that affects the nervous system
- D. Invasion of the gastrointestinal mucosa

7. What is the primary function of Protein A in *Staphylococcus aureus*?

- A. It binds to fibrinogen to promote clotting.
- B. It enhances phagocytosis by immune cells.
- C. It binds to the Fc region of antibodies, preventing opsonization.
- D. It produces toxins that lyse red blood cells.

8. Which antibiotic class is most commonly used in treating Methicillin-sensitive *Staphylococcus aureus* (MSSA) infections?

- A. Macrolides
- B. Tetracyclines
- C. Beta-lactams
- D. Fluoroquinolones

1. **Answer:** C. *Staphylococcus aureus*
2. **Answer:** D. Toxic shock syndrome toxin-1 (TSST-1)
3. **Answer:** B. Coagulase test
4. **Answer:** C. Device-associated infections
5. **Answer:** D. MRSA has an altered penicillin-binding protein (PBP2a) that reduces methicillin binding.
6. **Answer:** B. Production of a heat-stable enterotoxin in food
7. **Answer:** C. It binds to the Fc region of antibodies, preventing opsonization.
8. **Answer:** C. Beta-lactams

Further reading:

Jawetz, Melnick & Adelberg's Medical Microbiology, 26th edition-
Section 3: Bacteriology-
Chapter 13: The Staphylococci

Murray - Medical Microbiology 8th Edition
Section 4: Bacteriology
Chapter 18: STAPHYLOCOCCUS AND RELATED GRAM-
POSITIVE COCCI