PERIODONTICS

DENTISTRY2017-UNIVERSITY OF JORDAN

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Sheet   Slide

Etiology of periodontal diseases

- Our body contains 1.3 to 10 times more bacteria than human cells.
- Bacterial population comprises 2 kg of the total body weight (heavier than the brain which weighs about 1.4 kg)
- Oral bacterial microbiome of adults encompasses approximately 700 commonly occurring species.
- Most oral bacteria are harmless commensals under normal circumstances (they learned to live with us and we learned to live with them)
- Diseases only occur if there was imbalance between the host response and the amount of these bacteria and also the growth of different types of bacteria.

  in other words: disease occurs if increased mass and/or pathogenicity, suppression of commensal or beneficial bacteria, and/or reduced host response
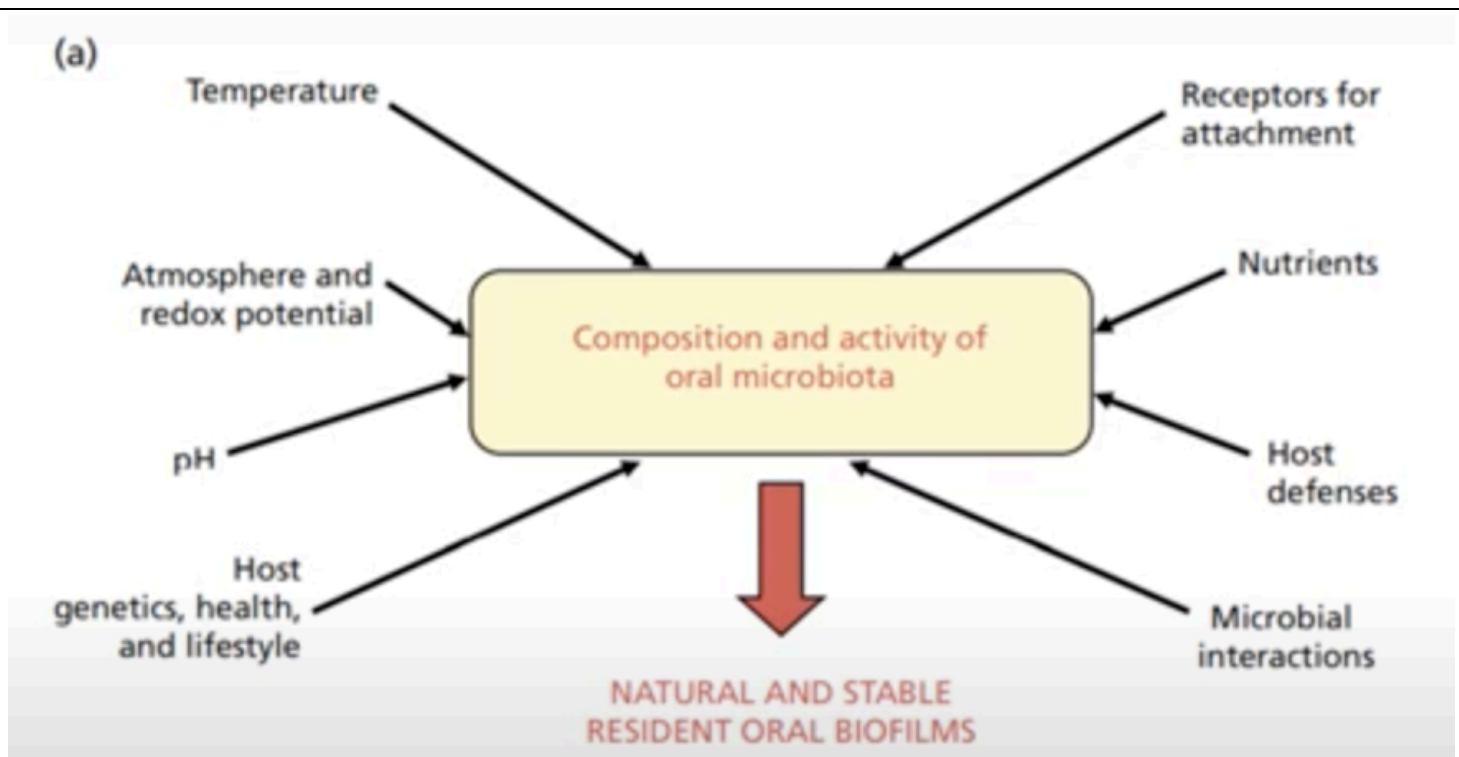
- Biofilms are composed of microbial cells encased within a matrix of extracellular polymeric substances, such as polysaccharides, proteins and nucleic acids so they live in an enclosed environment like a small city called actually slime city.
- It is very important for these microorganisms to stick around inside the oral cavity that they have some features that allow them to adhere to the oral cavity.
- Adherence on shedding or non-shedding surfaces

- So, adherence is an important feature to have a microflora inside the mouth, because as we know inside the mouth there is a lot of fluid movement, food movement, muscles movement also saliva, water and air so a lot of things are going around and if the microorganism doesn't have any feature that allows it to stick to the shedding or non-shedding surfaces it will pass into the lungs or GI tract.
- They usually stick to shedding surfaces like epithelial surfaces which usually change two times a day and the body cleans itself from mucosa surfaces but this doesn't happen with the non-shedding surfaces like the teeth for example.
- So, when brushing teeth don't brush the mucosa/ gingiva because they are covered with epithelium and epithelium already changes itself twice a day (shedding surface) but the problem is with the non-shedding surfaces like teeth, implants and prosthesis which should be cleaned.

Dental plaque: A host-associated biofilm consists mainly of a complex ecosystem of bacteria in addition to other microorganisms, forms clear or yellow soft deposits on teeth and other hard surfaces inside the oral cavity
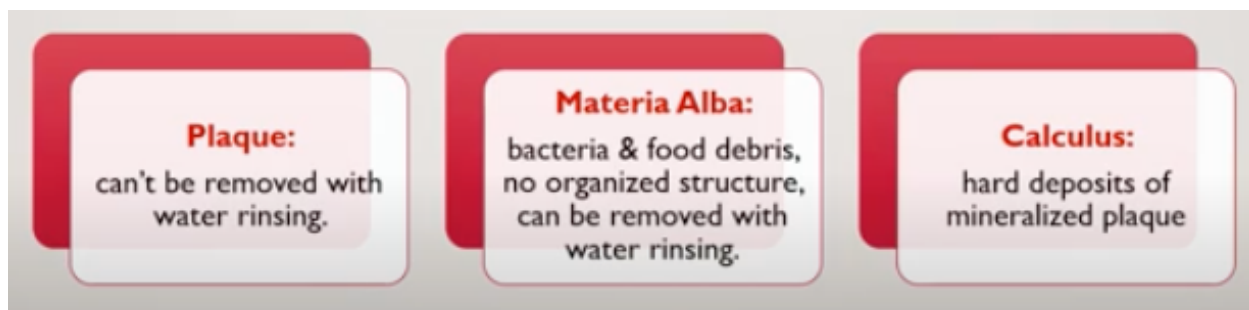
if you look at the background in the following figure, this an example of electron microscope photograph you can see different types and sizes of bacteria; rods and cocci around rods, spaces between cells, in addition to polysaccharides and proteins around them

DENTAL PLAQUE

- A host-associated biofilm
- Consists mainly of a complex ecosystem of bacteria
- Forms clear or yellow soft deposits on teeth and other hard surfaces

(a)

Temperature → Composition and activity of oral microbiota ← Receptors for attachment

Atmosphere and redox potential → ← Nutrients

pH → ← Host defenses

Host genetics, health, and lifestyle → ← Microbial interactions

NATURAL AND STABLE RESIDENT ORAL BIOFILMS

- composition and activity of oral microbiota depends on a lot of things mentioned in the picture above, all these things will lead eventually to a specific plaque.
- There is no plaque identical between different people and even between different areas inside the same mouth.

Tooth-accumulated materials

Plaque:
can't be removed with water rinsing.

Materia Alba:
bacteria & food debris, no organized structure, can be removed with water rinsing.

Calculus:
hard deposits of mineralized plaque

Clinically the biofilms inside the mouth can present as:

- plaque which can't be removed with water rinsing

- materia alba which is bacteria and food debris but it is not an organized structure and can be removed by water rinsing.

-calculus is hard deposits of mineralized plaque and can't be removed by water rinsing.

This is dental calculus that is firmly attached to the tooth

This is the materia alba that is a white collection of food debris and bacteria that stuck in between the embrasure and can be rinsed away with water

This is a 10 days old mature dental plaque (white creamy substance attached on the teeth) that already established an inflammation as we can see swelling and redness.

Microscopic structures of dental plaque:

1. Supra-gingival plaque: found at or above the gingival margin
2. Marginal plaque: supra-gingival plaque in direct contact with gingival margin
3. Sub-gingival plaque: found below the gingival margin (apical), between the tooth and gingival sulcular tissue. Sub-gingival plaque is what we are concerned about we are also concerned about supra-gingival plaque but for more severe disease sub-gingival plaque has tooth associated areas and tissue associated areas.

Composition of dental plaque:

1. Microorganisms:

The primary constituent of dental plaque is microorganisms

- Mainly Bacteria (the most common), yeast, protozoa, viruses and mycoplasma are also found in the dental plaque
- More than 700 species of bacteria in the mouth
- It forms 70% of the mass of dental plaque.

2. Intracellular matrix: polysaccharides, glycoproteins, lipids etc… that is produced either by bacteria or by the host.

3. Host cells.

Intra-cellular matrix:

- 20-30% of plaque
- organic constituents:
  a. glycoproteins: from saliva, the bacteria use it to start to attach on the tooth.
  b. mucopolysaccharides: bacteria (dextran), the bacteria make their own polysaccharides
  c. proteins: from GCF (albumin) which already come from blood
  d. lipids: come from disrupted bacterial and host cells

- inorganic constituents:
  mainly calcium and phosphorus (you can find sodium, magnesium and fluoride at much lower levels) which with time it gets more mineralized and turn into calculus.

 source: saliva (supra-gingival plaque) or GCF (subgingival plaque)
  Why is this important? Both supra-gingival and sub-gingival plaque will become mineralized and hardened turning into calculus and since each one has a different source of mineralization there will be difference between them.
  As you can see in the picture this is an extracted tooth due to periodontal disease, notice the very dark brown/ black layer of calculus (sub-gingival calculus) which is usually harder and more tenacious (difficult

to be removed). Its color is black because the source of mineralization is the blood that is in the plasma that comes with the GCF so it gets part of the color of the blood.

- Supra gingival calculus is lighter white-yellow in color and is brittle because the source of the calcium and phosphate is actually from the saliva.
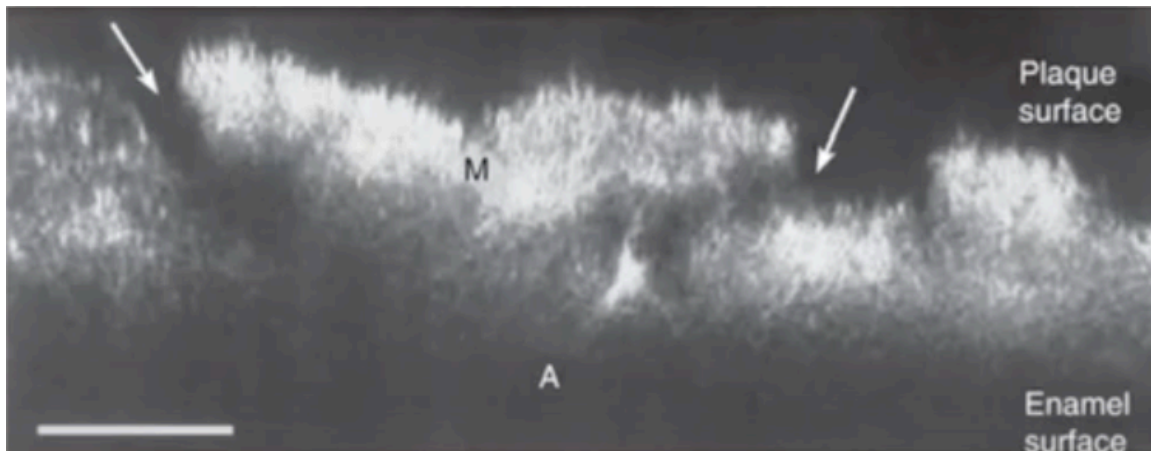
If you look at this electron microscope photograph you can see that this is a dental plaque section and the white arrows point on fluid filled channels running through the plaque mass, it is like a city and those are the roads to transform bacteria and nutrients between the residents of this city.

-heterogeneous in structure.

Formation

- Bacteria is detectable within 3 minutes starting from the gingival third so we can actually see plaque within 3 minutes but that doesn't mean we have to keep brushing our teeth all the time.
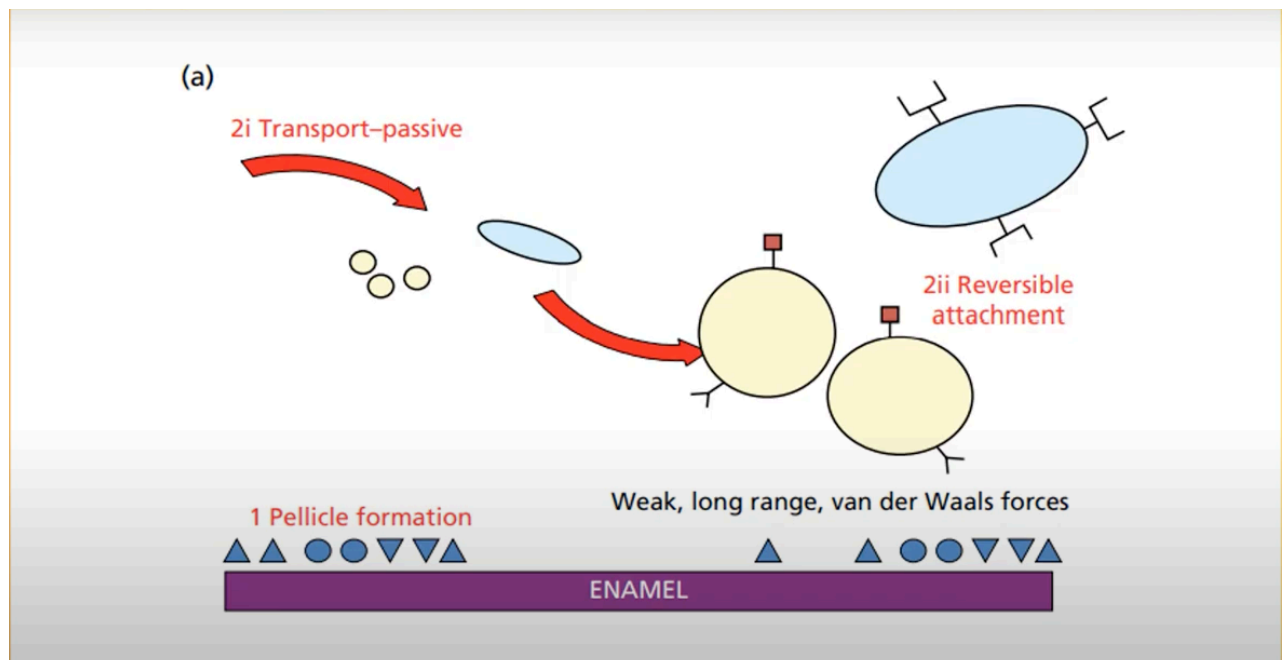
Stages:

1. Acquired pellicle
2. G +ve facultative Anaerobes (they usually use oxygen if available and if not available they do fermentation so they can work as aerobes or anaerobes)
3. G –ve Anaerobes

-After this pellicle form all the other bacteria start aggregating, proliferating and maturing into a plaque mass

-The pellicle is formed from glycoprotein and has two layers, the basal layer was formed at the time when the teeth eruption started and it can't be removed we can just remove the superficial layer (while brushing teeth) and it forms again within one minute.

So dental enamel is permanently covered with an acquired pellicle from the moment that teeth erupt.

Schematic representation of the different stages in the formation of a biofilm
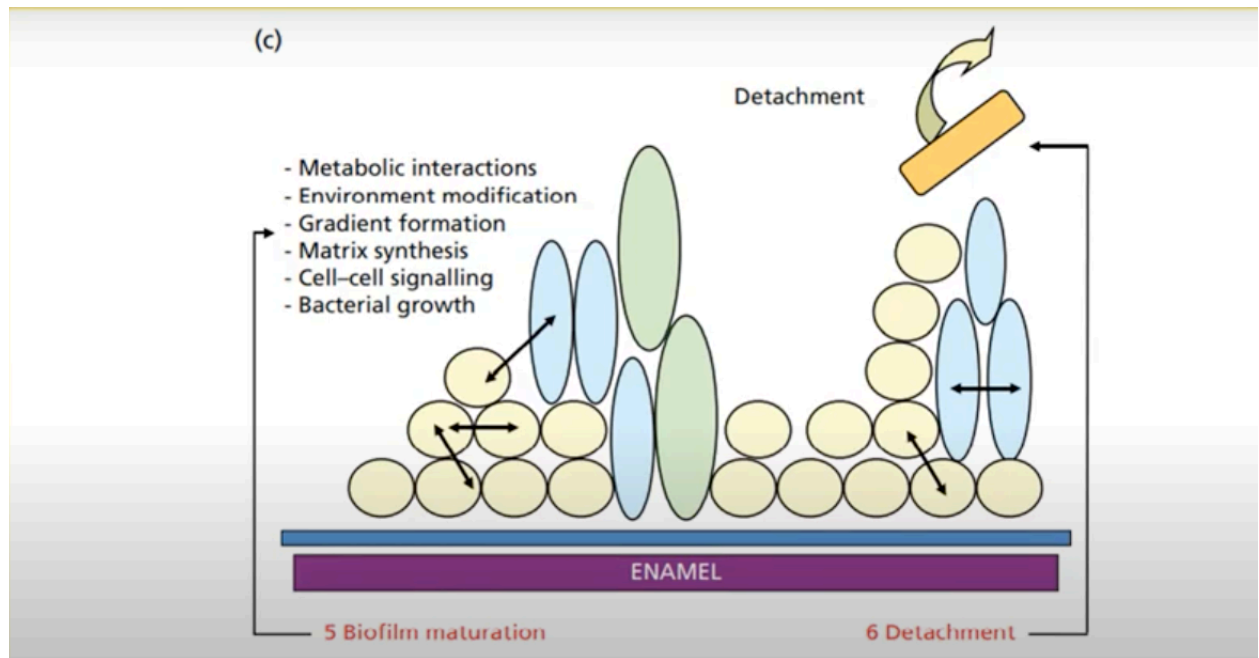
First stage this is the pellicle formation on enamel, after you brush your teeth pellicle starts forming. Then the bacteria come close to the surface of enamel usually by passive transport (chewing, moving your teeth, saliva, fluid). But what gets it closer to the pellicle and enamel is the van der waals forces (weak attachment, long range).
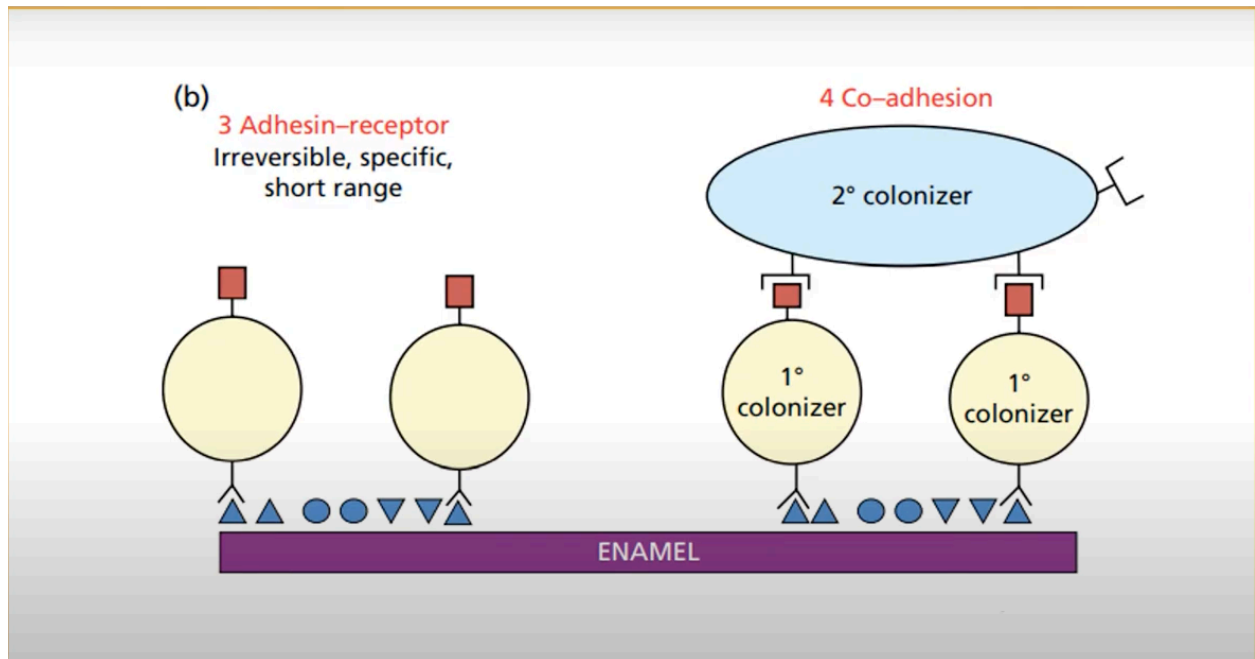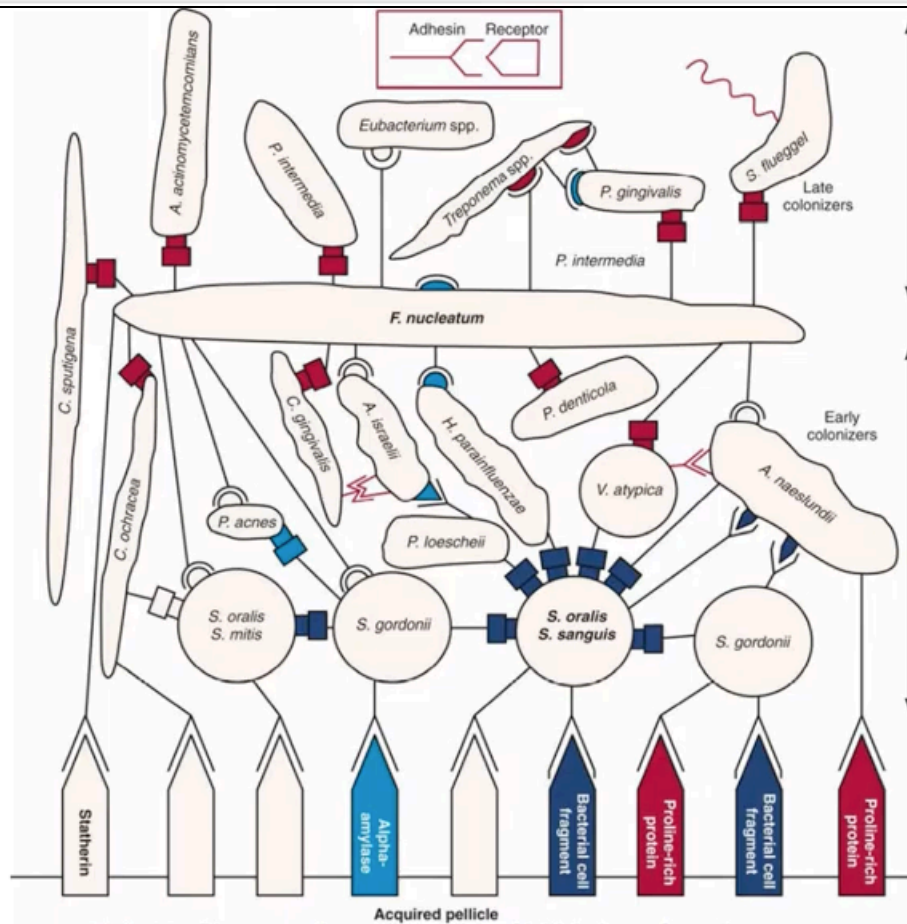

The strong attachment is achieved when the adhesives which is found on the bacteria attaches to certain shapes, bacteria or charges (positive charge attaches with negative) found on pellicle forming the first colonizer (streptococcus in the picture). These have their own receptors which attach to the secondary colonizer (rods in this picture) by co-adhesion.

After the co-adhesion there is something called co-aggregation where the bacteria starts getting packed next to each other as we will see in the next photograph

(c)

Detachment

- Metabolic interactions
- Environment modification
- Gradient formation
- Matrix synthesis
- Cell–cell signalling
- Bacterial growth

ENAMEL

5 Biofilm maturation   6 Detachment

A quick revision of the stages as they were mentioned in the slides: (regarding the number of the stages written on the pictures 1-6)

1. Pellicle forms on a clean tooth surface

2i. bacteria are transported passively to the tooth surfaces

2ii. Where they may be held reversibly by weak, long-range forces of attraction

3. Attachment becomes more permanent through specific stereochemical molecular interactions between adhesins on the bacterium and complementary receptors in the pellicle.

4. Secondary colonizers attach to the already attached primary colonizers by molecular interactions (co-adhesion)

5. Growth results in biofilm maturation, facilitating a wide range of intermicrobial interactions (synergistic and antagonistic)

6. On occasions, cells can detach to colonize elsewhere

Diagram showing the arrangement of initial dental plaque formation. Labels include: Adhesin, Receptor; Eubacterium spp.; A. actinomycetemcomitans; P. intermedia; Treponema spp.; P. gingivalis; S. flueggei; Late colonizers; P. intermedia; F. nucleatum; C. sputigena; C. gingivalis; A. israelii; H. parainfluenzae; P. denticola; Early colonizers; C. ochracea; V. atypica; A. naeslundii; P. acnes; P. loescheii; S. oralis, S. mitis; S. gordonii; S. oralis, S. sanguis; S. gordonii; Statherin; Alpha-amylase; Bacterial cell fragment; Proline-rich protein; Bacterial cell fragment; Proline-rich protein; Acquired pellicle.

In this diagram, we can see the pellicle and the first colonizers which are all streptococci (gram positive aerobic bacteria) (S. oralis and S. sanguis are the most common) and they are the most dominant, these first colonizers by co-adhesion they attach to the secondary colonizers. It is a complex network of attachment. We also have bridging organisms (bacteria) which have multiple receptor sites so it helps in building a block for building a much larger mature plaque.

- Fusobacterium nucleatum is an example of a bridging bacteria.
- The bacteria from the early colonizers that give you the late colonizers are the worst bacteria which are P. gingivalis, Treponema spp., P. intermedia and A. actinomycetemcomitans

- These four species are important because they are the nastiest periodontal pathogens.
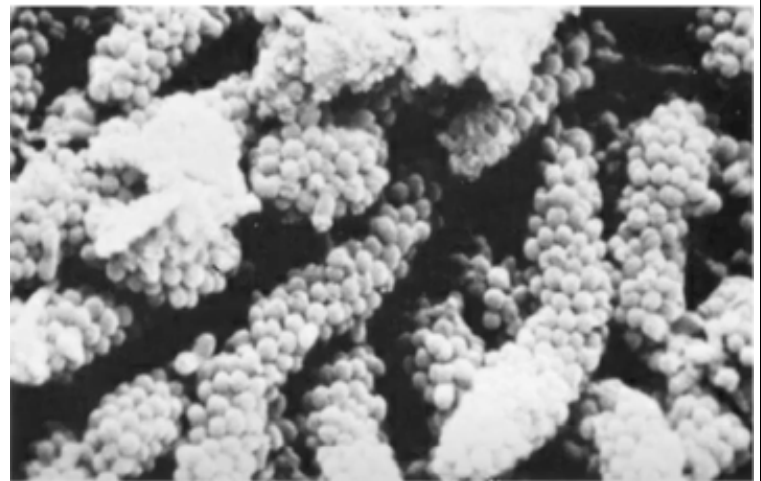
Supra-gingival plaque:

When we look at mature supra-gingival plaque we see stratified layers of different bacteria

At the tooth surface we find the G+ve cocci and short rods near the surface because they are the most common and always available next to the tooth

When the plaque become mature, on the outer surface of the mature plaque mass we will see G-ve rods, filaments and spirochetes.

Long-standing supragingival plaque near the gingival margin demonstrating a 'corn cob' arrangement. A central gram negative filamentous core supports the outer coccal cells, which are firmly attached by interbacterial adherence or coaggregation. (these are rods with cocci attached to them)
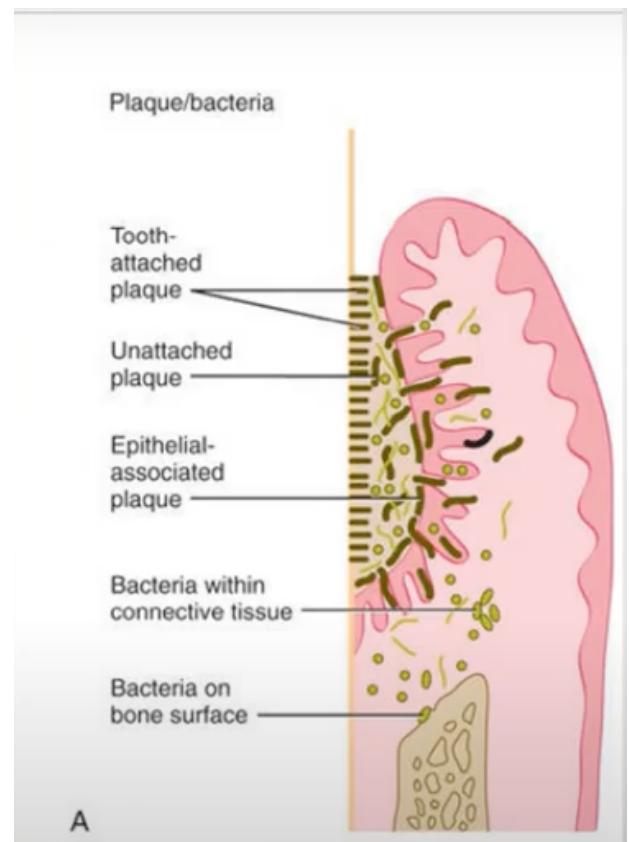
Sub-gingival plaque:

It is different and has different types of bacteria because it has different types of nutrients like blood products and more proteins and it is hidden outside the oral cavity so it favors anaerobic environment especially if you go deeper inside the pocket (the more deeper you go the more anaerobic bacteria you find)

- Tooth associated region: G+ve rods and cocci
- Tissue associated region: G-ve rods and cocci, spirochetes

This diagram shows the tooth associated region where the plaque is more organized. But in the tissue associated region you can see that the bacteria are leaving the plaque layer and they become free bacteria and they invade the tissue, what invades first is the motile bacteria (spirochetes) we can find them inside the tissue and sometimes on the surface of the bone

Plaque/bacteria

Tooth-attached plaque

Unattached plaque

Epithelial-associated plaque

Bacteria within connective tissue

Bacteria on bone surface

A

- An analysis of more than 13000 plaque samples that looked for 40 subgingival microorganisms using a DNA-hybridization 568 methodology defined color-coded "complexes" of periodontal microorganisms that tend to be found together in health or disease.
- The composition of the different complexes was based on the frequency with which different clusters of microorganisms were recovered, and the complexes were color coded.
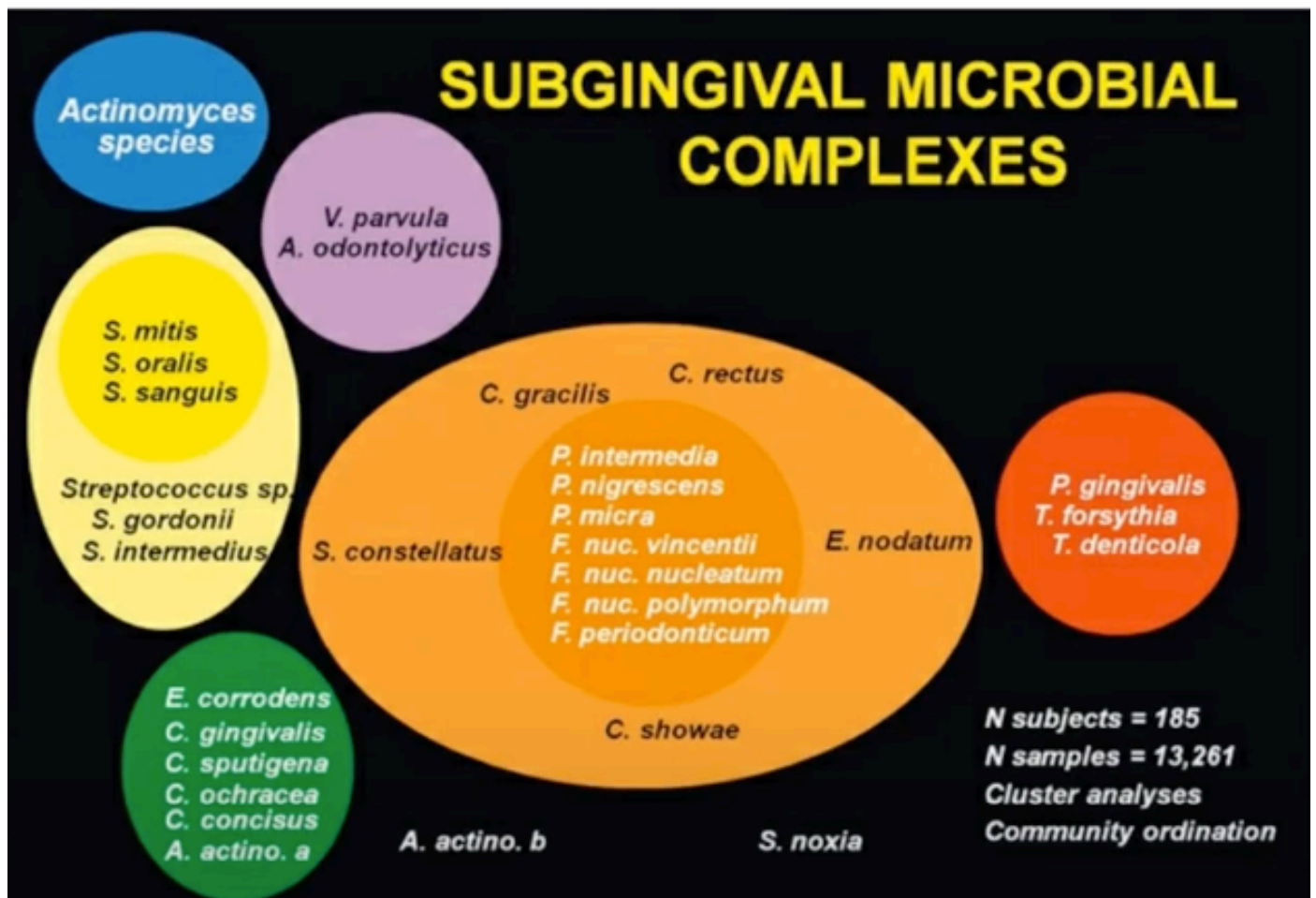
- The complexes to the left consists of species that are thought to colonize the tooth surface and to proliferate at an early stage. (yellow and purple).
- The streptococcus species (yellow) together with the purple species are called the early colonizers.
- The orange complex is big because it has a lot of types of bacteria, it is the bridging complex we find the bridging bacteria which bridge the early colonizers to the nasty red complex
- The orange complex becomes numerically dominant later; it is thought to bridge the early colonizers and the red complex species, which become numerically more dominant during the later stages of plaque development.
- The red complex consists of P. gingivalis, T. forsythia and T. denticola, these are associated with severe attachment loss and periodontitis.
- The green complex is considered a part of the bridging or early colonizers maxes a way for the bad red complex.

Plaque and Periodontal Disease

-The site specificity of plaque is significantly associated with different diseases of the periodontium.

-Plaque that is on the gingival margin (marginal plaque) is associated with **gingivitis**, especially if the patient doesn't clean well and there are other factors present.

-Supragingival plaque and tooth-associated subgingival plaque (باتجاه السن apical) are critical in **calculus formation and root caries**.

-Tissue-associated subgingival plaque is important in the tissue destruction that characterizes different forms of **periodontitis**.

-Biofilms can stick on prostheses and surface of implants and cause **peri-implantitis** and **peri-implant mucositis** (we'll take them in future lectures).


The Biofilm Environment

By now, we know that biofilm contains different types of bacteria, whether supragingival or subgingival and we discussed the differences between both, and we talked a bit about the types of bacteria and the subgingival microbial complexes. Now, what happens inside the biofilm?

The environment inside the slime city (biofilm) is complicated, there is a **physiologic interaction** between the different constituents. They communicate with each other. An example of **communication** is something we call the Quorum sensing (QS).

Quorum sensing: Scientists believe that when bacteria first start accumulating, they release certain peptides. After a while and when these peptides reach a certain concentration, the bacteria now knows that it's not alone (it reached a certain number) that it can actually start producing and cooperating together. So now, they can attack other bacteria or promote the growth of beneficial bacteria that helps them and gives them the nutrients they need. This is how they communicate.

Further information about Quorum sensing (not required/ from multiple sites on the internet/you can skip if you like):
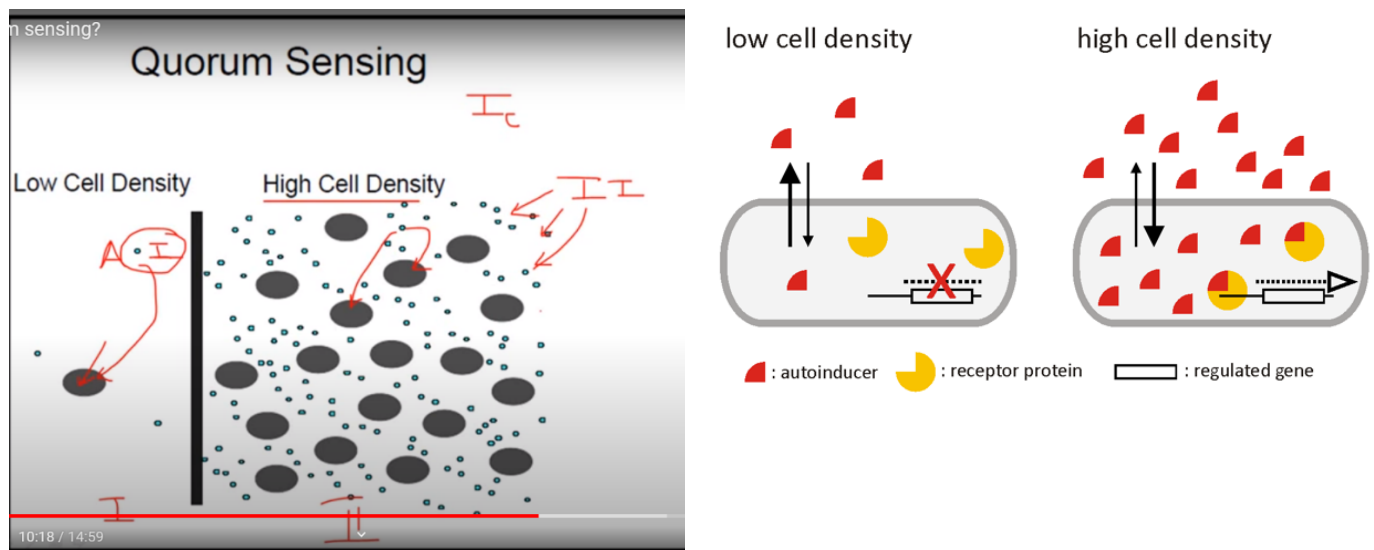
Quorum sensing is a mechanism of communication between bacterial cells (microorganisms) via **chemical signaling**.

How does quorum sensing work? (check with the pictures above) During their reproductive cycle, individual bacterium synthesize and release chemical signal molecules called **autoinducers**. When these autoinducers reenter the bacterial cell, a set of different proteins can be activated producing a certain effect. **So, autoinducers alter gene**

expression generating a certain effect. But for that to happen the bacterial colony needs a certain concentration of autoinducers. When we have a few number of bacteria only (low cell density), the autoinducers produced will not be enough to trigger the desired effect (process). As the bacteria continue to reproduce, the number of bacterial cells increases (cell density increases), and the extracellular concentration of the autoinducers increases as well. Finally, when the autoinducers reach a certain concentration threshold, they lead to an alteration in gene expression.

Why do bacteria use quorum sensing? Quorum sensing allows bacteria populations to communicate and coordinate **group behaviour**. It allows them to regulate various processes such as virulence, horizontal gene transfer, biofilm formation, and competence.

Google definition: Quorum sensing is the regulation of gene expression in response to fluctuations in cell-population density.

Cell-population density increases> concentration of autoinducers increases> alteration (regulation) of gene expression > coordination of group behavior.

———————————————

Let's continue,

-What is the importance of this communication?

Sometimes, this communication makes the biofilm nastier or more resistant to antibiotics.

-Susceptibility to antibiotics: bacteria that are found within biofilms are 1000 times more resistant to antimicrobial agents than their planktonic (free bacteria that is not attached to a dental plaque mass) counterparts.

-How would biofilms increase or reduce susceptibility to antibiotics?

Either by:

✓ If some bacteria are already resistant to this antimicrobial, then they will **exchange resistant genes by conjugation** (transfer of genetic material between bacterial cells).

✓ Also, if the drug targets the cell membrane to burst the bacterial cell, there are **multidrug resistance pumps** found on the cell wall of bacteria. So, when this drug (antibiotic) comes to burst the cell, the bacteria absorbs it and gets it out through these pumps.

✓ Certain bacteria uses **extracellular enzymes such as β-lactamases**, that cleave the β-lactam ring in penicillin so they become penicillin resistant.
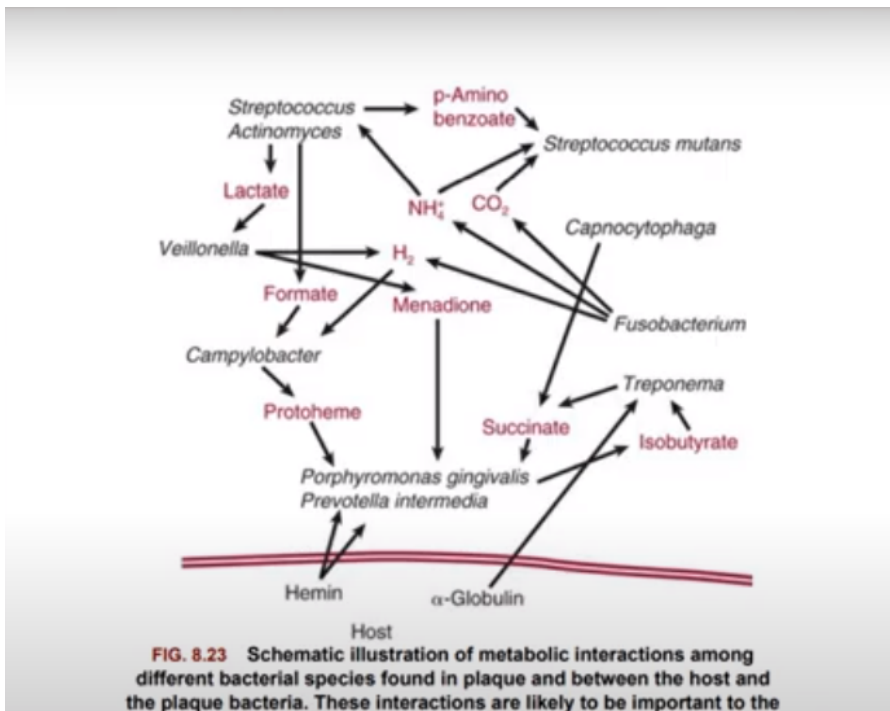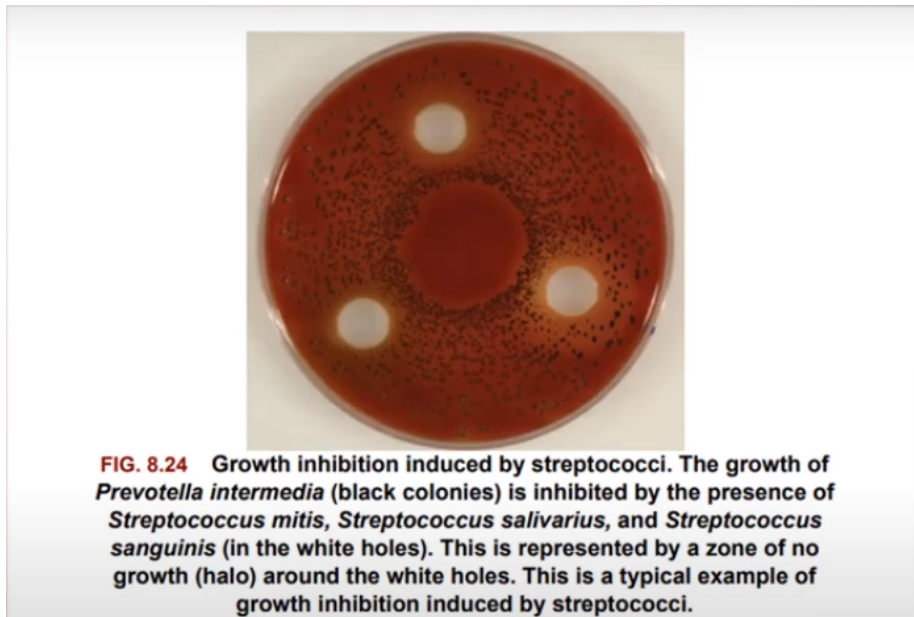
➢ This is a schematic illustration of metabolic interactions among different bacterial species, it shows you what each type of bacteria produces and how it benefits another type and so on.

FIG. 8.23 Schematic illustration of metabolic interactions among different bacterial species found in plaque and between the host and the plaque bacteria. These interactions are likely to be important to the

Streptococcus and Periopathogens

-The interesting thing about bacteria is that it's not always rainbows and butterflies inside the biofilm. Like us, sometimes there are problems between neighbours.

-There are clinical studies that state that S. sanguinis and some periodontal pathogens have **antagonistic relationships** (if one increases in number, the other will decrease).

FIG. 8.24   Growth inhibition induced by streptococci. The growth of *Prevotella intermedia* (black colonies) is inhibited by the presence of *Streptococcus mitis, Streptococcus salivarius,* and *Streptococcus sanguinis* (in the white holes). This is represented by a zone of no growth (halo) around the white holes. This is a typical example of growth inhibition induced by streptococci.

> ➤ In this example, **Prevotella intermedia** (one of the bacteria identified to be related to periodontal disease) which is represented by the black dots, is growing. But, in the zones where there are streptococcus species, we can see an empty area (white halo) because it's growth is inhibited.

-Lab studies have shown that **competitive interaction exists between streptococci and periodontopathogen**. e.g. S.mitis have been shown to inhbit hard and soft tissue colonization of P.gingivalis. So, sometimes they compete for colonizing the tissue (for space).

Biofilm Nasty Residents

Other than bacteria, there are different types of animals and residents in the biofilm. (viruses, fungi, protozoa)

As we believe and as studies are showing recently, sole bacterial-host interactions appear insufficient to explain the clinical characterisics of the disease. For example, it's not as simple as measles where 'this virus causes this problem, and if you give them this vaccine or treatment it will be healed'. It's not as simple as that.

- **Viruses**: the hypothesis about herpesviruses and its etiology in periodontitis. Some studies say that it has an etiological role and some studies say that it doesn't, so it's still controversial.
- Same thing for Candida **(Fungi)**, and
- **Protozoa**: Entamoeba gingivalis, Trichomonas tenax

Dr. Omar was recently involved in a study conducted on JU dental students' patients, in which they took samples from patients with severe, and mild periodontitis and gingivtitis. And they actually saw the entamoeba gingivalis (protozoa) swimming inside the plaque samples, like worms living inside the plaque (protozoa are larger microorganisms).
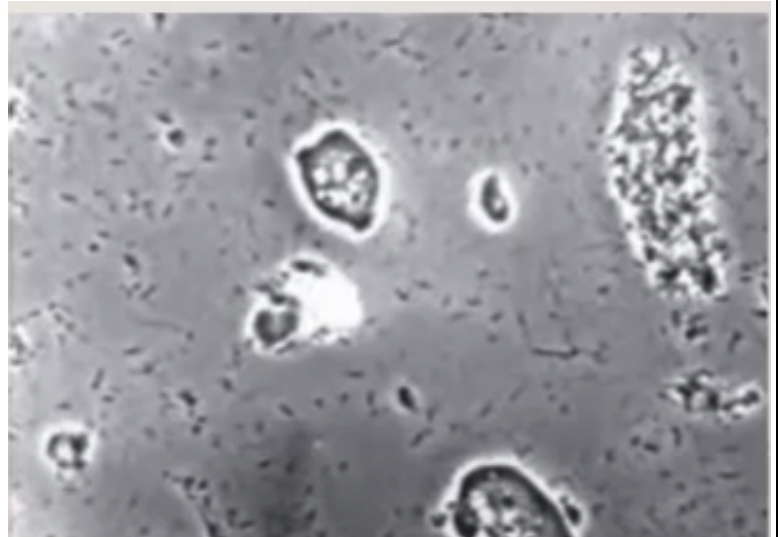
So, it's interesting to know that it's not only bacteria. Probably, there are other organisms involved in the pathogenesis of periodontitis.

Microbial Specificity of Periodontal Disease

How did we reach the conclusion that we know now that plaque causes gingivitis? Is it the accumulation of large or thick plaque masses (quantity of plaque), or is it about the type of bacteria inside the plaque or what is it about really?

We have 3 hypotheses:

- Non-specific plaque hypothesis
- Specific plaque hypothesis
- Ecologic plaque hypothesis

I. <u>Non-specific plaque hypothesis:</u>

- What is important is: *How many bacteria is there?* (quantity)

-The concept is that all bacteria are equally pathogenic, but if it increased in number disease happens. So,what's important is the number of bacteria, not its type. If the bacteria accumulated and the plaque became thick, it means you're going to get periodontal disease.

-This hypothesis was disregarded ×, because:

- Some people had very severe inflammation and destruction, but when you look at their mouth there is barely biofilm. "levels of plaque inconsistent with levels of inflammation in some patients".
- Also, in some people you find plaque and calculus all over the mouth, but you see periodontitis and attachment loss in few sites not all sites. "site specificity of periodontal lesions"

II. <u>Specific plaque hypothesis:</u>

-After that they said that it's not about the quantity, it's about the quality (type) of bacteria present. *Which bacteria?*

-If you have the nasty microorganisms, then you will have periodontitis or problems whether there is a lot of bacteria or not.

-So, they depended on the pathogenicity of certain nasty microorganisms. "pathogenicity of dental plaque depends on the presence of or an increase in specific microorganisms".

-This hypothesis was supported when they recognized the presence of **A. actinomycetemcomitans (Aa),** especially in paients that have very severe disease and that are young.

-So, they said that this bacteria is the cause of very severe and early attachment loss. However, after studies they revealed at the end that:

- Some people have this bacteria, but they don't really have periodontal disease.
- While other people don't have this bacteria, but they have periodontal disease.

-Eventually, this hypothesis was disregarded× as well.

- Disregarded
  - ✓ studies do not reveal whether the presence of specific bacteria causes or correlates with the presence of disease.
  - ✓ studies have shown that periodontal disease can occur even in the absence of defined "pathogens," such as red complex bacteria, and conversely that "pathogens" may be present in the absence of disease.

I.   Ecological plaque hypothesis:

-In the 90's, Marsh and his collegues suggested the ecological plaque hypothesis. Where we don't only look at the plaque, it's quantity or type. Instead, we look at the overall plaque and the environment surrounding it.

-We have certain numbers and types of bacteria that are already living inside our oral cavity in a state of **microbial homeostasis** (there is balance between the host, environment and the bacteria inside the biofilm).
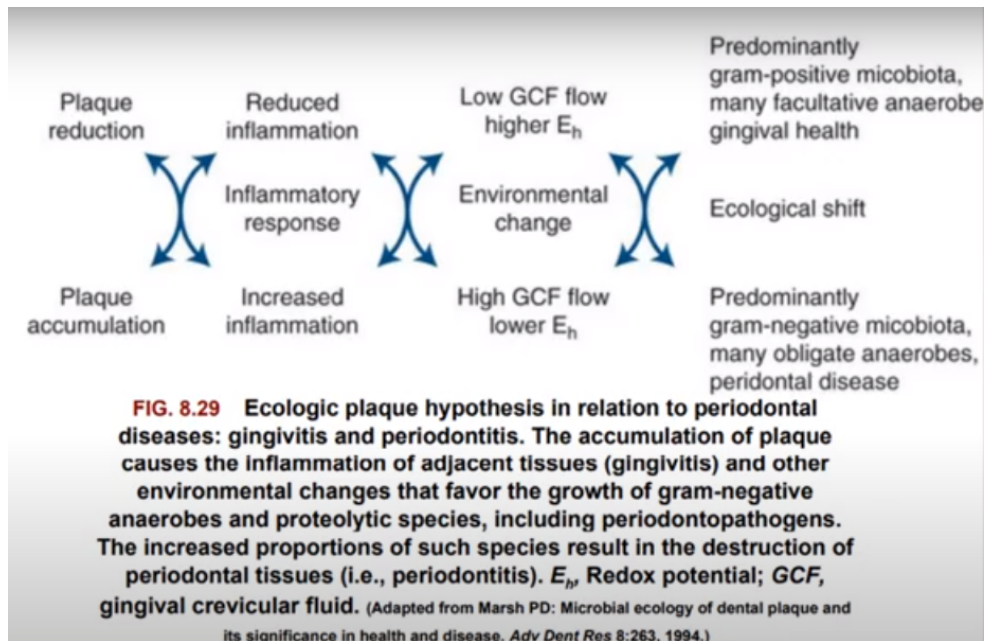
-This homeostasis can shift to **dysbiosis**, where the environment will no longer be favorable to the good bacteria, rather it will be more favorable to the bad bacteria.

-Why does this happen? What is the etiology? Usually it's microbial, host, or environmental changes that will cause dysbiosis.

-How do we treat it? Treating the plaque only doesn't always solve the problem. Sometimes, we give antibiotics or make other interventions. Not only to remove the microorganisms, sometimes we need to improve the host immunity, or stop them from smoking for example or stop their diabetes etc.

-You need to control the **microbial, host, and environmental** factors to restore microbial homeostasis.

"Targeting speficic microorganisms may be less effective because the conditions for disease will remain".

FIG. 8.29 Ecologic plaque hypothesis in relation to periodontal diseases: gingivitis and periodontitis. The accumulation of plaque causes the inflammation of adjacent tissues (gingivitis) and other environmental changes that favor the growth of gram-negative anaerobes and proteolytic species, including periodontopathogens. The increased proportions of such species result in the destruction of periodontal tissues (i.e., periodontitis). E_h, Redox potential; GCF, gingival crevicular fluid. (Adapted from Marsh PD: Microbial ecology of dental plaque and its significance in health and disease. Adv Dent Res 8:263, 1994.)

➤ This diagram shows you the ecologic plaque hypothesis. Plaque accumulation is associated with increased inflammation, also if there was environmental changes this will lead to ecological shift (you will have more gram-negative bacteria, anaerobic bacteria etc.) especially if there was bad immunity, you're going to have the disease. So, this is more realistic to believe in.

Keystone Pathogen Hypothesis and Polymicrobial Synergy and Dysbiosis Model

-Recently, there is a new theory that states that there is a *keystone pathogen*. Which means that what we said earlier is correct, but still there are pathogens when present, even in low concentration, that can orchestrate inflammatory disease by remodeling a normally benign bacteria into a dysbiotic (nasty) one.

-They release certain enzymes, proteases, or products. And this causes the bacteria around it to become nastier. So, they make a revolution against the body and start periodontal disease.

-An example of it is **<u>P.gingivalis</u>**, it subverts the host immune system and changes the microbial composition of dental plaque despite being less than 1% of total mirobiota.

-This keystone pathogen is associated with microbial synergy and accessory pathogens (pathogens that change their attitude and become nastier and more pathogenic), and this leads to a state of dysbiosis.

Role of Beneficial Bacteria

We know that probiotics are beneficial for the gut. Sometimes, they even treat some gastrointestinal tract diseases with probiotics. So, some theories state that you need to use them to treat periodontal disease and decrease the amount of nasty plaque. How? We already talked about the example of how streptococcus affected the colonization of some periodontopathogens such as P.ginivalis (page 21).

These bacteria (beneficial bacteria) can affect the pathogenic species in different ways and thus modify the disease process as follows:

- by passively occupying a niche (space) that may otherwise be colonized by pathogens,

- by actively limiting a pathogen's ability to adhere to appropriate tissue surfaces (inhibit its colonization)

- by adversely affecting the vitality or growth of a pathogen

- by affecting the ability of a pathogen to produce virulence factors

- by degrading the virulence factors produced by the pathogen. (for example if this pathogen has a certain virulence factor such as an enzyme that kills WBC's, the beneficial bacteria can do something

to counteract this enzyme or to kill this pathogen).E.g.the effect of S.sanguinis that produces H2O2, which either directly or by host enzyme amplification can kill A.actinomycetemcomitans (nasty bacteria).

Virulence Factors of Periodontal Microorganisms

-What makes this bacteria more pathogenic and nasty compared to other bacteria?

It needs to attach to surfaces, invade tissues, and evade the host immune response.

-Can be subdivided as follows:

1-factors that promote colonization (adhesins)

2-toxins and enzymes that degrade host tissues, and

3-mechanisms that protect pathogenic bacteria from the host.

Example: **P.gingivalis**

1.Strains of P.gingivalis produce two types of fimbriae (like hands for the bacteria), which are known as the major fimbriae and the minor fimbria that allow it to attach and get inside the epithelial cells, so it invades the body tissue and causes attachment loss. (the host also plays a role in causing attachment loss, but we'll discuss that later with Dr.Ahmad).

2. gingivalis gingipains are proteases (an enzyme that cleaves protein, collagen is a protein and it's the main constituent of PDL).

3. Also, they can evade the host response or the phagocyte can't recognize that it's a weird body, or they can degrade the complement system that is responsible about bursting bacterial cells. So, another

bacteria might enter the body because P.gingivalis made gingipains that can degrade complement components C3,C4,C5.

Evidence Supporting a Role for *A. actinomycetemcomitans* and *P. gingivalis* as Pathogens in Periodontal Disease: Socransky's Criteria

Criterion	A. actinomycetemcomitans	P. gingivalis
Association	Increased in localized aggressive periodontitis (LAP) lesions Increased in some chronic periodontitis lesions Detected in the tissues of LAP lesions	Increased in periodontitis lesions Found associated with crevicular epithelium
Elimination	Suppressed or eliminated in successful therapy Found in recurrent lesions	Suppressed or eliminated in successful therapy Found in recurrent lesions
Host response	Increased serum and local antibody levels in LAP	Increased systemic and local antibody levels in periodontitis
Animal studies	Capable of inducing disease in gnotobiotic rats	Found to be important in experimental mixed infections and in periodontitis in the cynomolgus monkey
Virulence factors	Host tissue cell invasion, leukotoxin, collagenase, endotoxin (LPS), epitheliotoxin, fibroblast inhibiting factor, bone resorption-inducing factor	Host tissue cell adherence and invasion, collagenase, trypsin-like enzyme, fibrinolysin, phospholipase A, phosphatases, endotoxin (LPS), H_2S, NH_3, fatty acids, factors that affect PMN function

➢ This table shows you the virulence factors that are found in (Aa) and (P.gingivalis)
  Leukotoxin is a toxin that kills leukocytes
  Collagenase breaks collagen
  *The doctor only mentioned what's underlined (but read all the factors in case)

Microbiology of Specific Periodontal Conditions

-**Koch's postulate:** has certain criteria to be applied, not all are applicable in periopathogens.

-So, there is **Socransky's criteria** (more recent), which states that:

For a Perio pathogen to be a pathogen ,not commensal bacteria from normal microflora, it must:

1-be present in increased numbers in diseased sites

2-decreased numbers in clincal resolution (after you treat the patient it should decrease in number)

3-induce an immune response
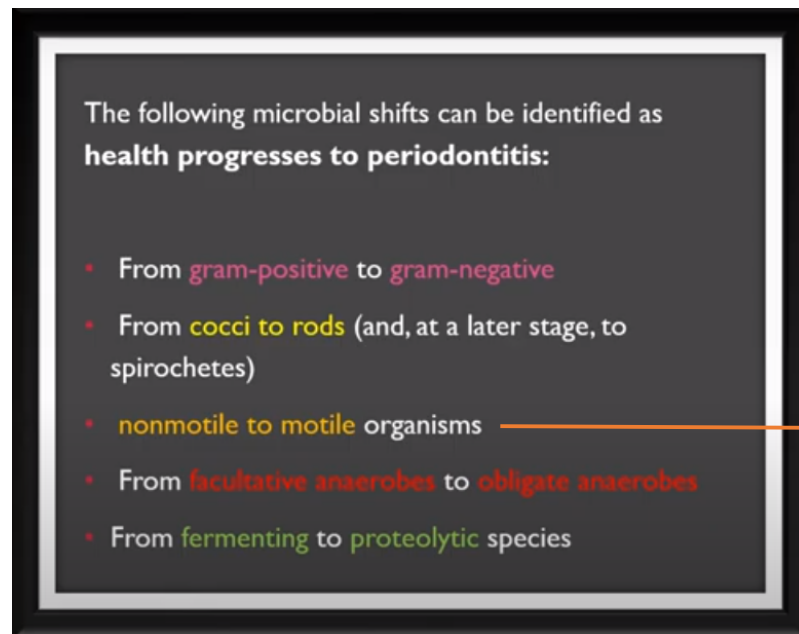
4-cause disease in experimental animal models

5-demostrate virulence factors

For example, P.gingivalis if it was observed that in disease it was increasing in number and after treatment it decreases, and it has virulence factors that induce disease, and also it causes disease in animals, and it's immunogenic (produces an immune response). Then, it's a perio pathogen.

Health-Gingivitis-Periodontitis:

Now, how will what we learned affect us clinically?

What is the microbial situation that is present in health, in gingivitis, and in periodontitis?

The following microbial shifts can be identified as **health progresses to periodontitis:**

- From gram-positive to gram-negative
- From cocci to rods (and, at a later stage, to spirochetes)
- nonmotile to motile organisms → It becomes motile, to invade the tissues.
- From facultative anaerobes to obligate anaerobes
- From fermenting to proteolytic species

These are the changes that identify shift of microorganisms into nastier ones. (from health to periodontitis)

**So you might be asked the following question: What are the microorganisms that are present in the state of health?

You have to suppose that it's probably more G+ve, cocci, there might be some rods but it will be G+ve rods not G-ve, nonmotile bacteria, facultative anaerobes, and fermenting species.

And, this is exactly what they found in the studies:

Periodontal health:

-Primarily G+ve Facultative species

-Streptococcus and Actinomyces genera (primary colonizers)

-Small proportions of G-ve species

Gingivitis: in between

-Initially> G+ve cocci and rods and some G-ve cocci

-Later> G-ve rods (with more mature plaque and more advanced gingivitis)

-Eventually, equal proportions of G+ve and G-ve (50/50 or 45/55 %)

Chronic periodontitis:

-Anaerobic (90%)
-G-ve bacteria (75%)

- With a lot of Spirochetes (motile microorganisms)

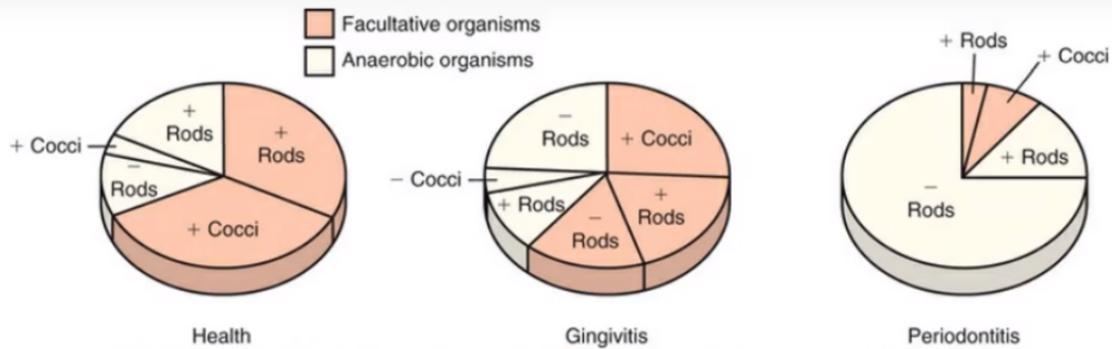-Pg (P.gingivalis), Tf (tannerella forsythia), Td (treponema denticola), 'red complex' Cr, Pi, Ec, Aa, Fn, Pm

Health   Gingivitis   Periodontitis

FIG. 8.31 **Pie charts based on culturing studies that represent the relative proportion of different morphotypes in subgingival samples in cases of periodontal health, gingivitis, and periodontitis. A clear distinction is made between facultative species and obligate anaerobic species. Spirochetes are not included. +, Gram-positive; −, gram-negative.**

➢ This pie chart shows you that in health you have more facultative organisms, while in gingivitis it decreases a bit, and in periodontitis it's mainly anaerobic. Regarding the shape, at the beginning we have cocci then it shifts more into rods, and in periodontitis it's mostly rods. As well, in health, it's mostly G+ve, almost 50/50 in gingivitis, and in periodontitis it's predominantly (75%) G-ve.

Micrographs of plaque samples:

A. Healthy patient: Cocci with some rods
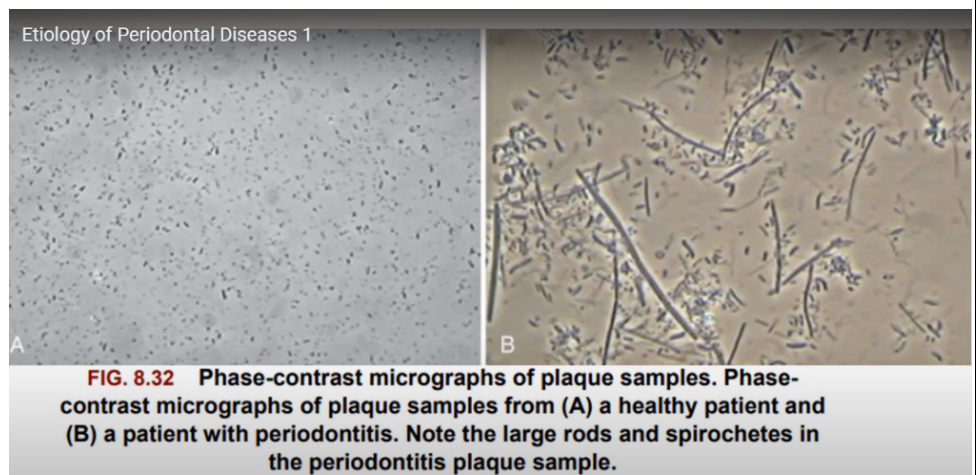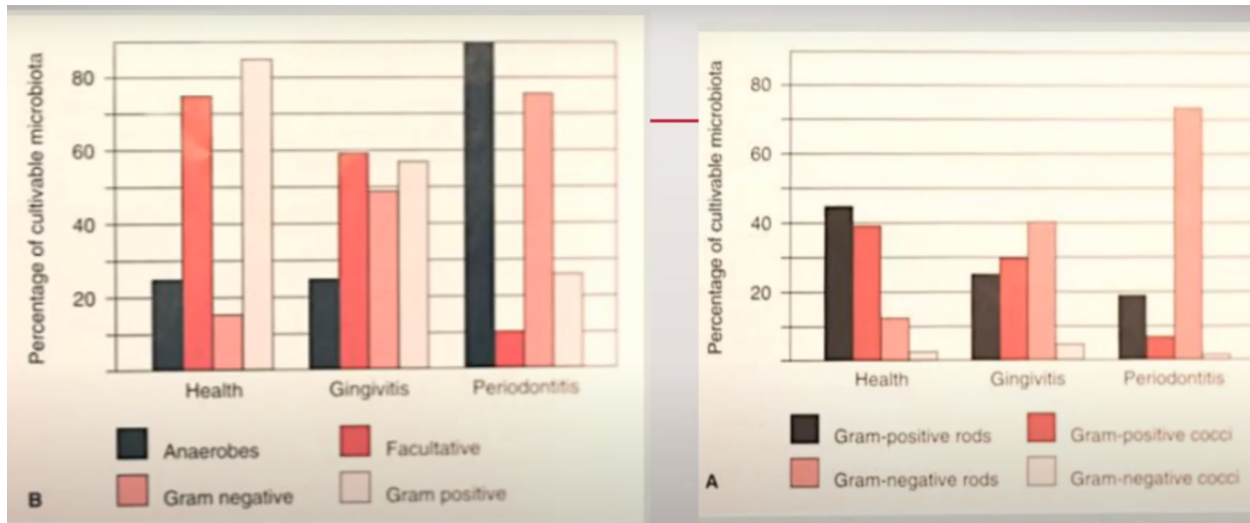B. Patient with Periodontitis: Mainly rods ,some cocci

FIG. 8.32 **Phase-contrast micrographs of plaque samples. Phase-contrast micrographs of plaque samples from (A) a healthy patient and (B) a patient with periodontitis. Note the large rods and spirochetes in the periodontitis plaque sample.**

B.In health the anaerobic bacteria was 20%, in gingivitis the same thing. While in periodontitis it shoots up to 90%. Whereas, the G+ve was the highest in health. It decreased to become almost equal to the G-ve in gingivitis. While in periodontitis, there is a huge difference (more G-ve).

A. G+ve rods was high in health, then it decreased a bit. Whereas the G-ve rods, from health to gingivitis to periodontitis, it shoots up to around 70%.
Anything G+ve is decreasing, while anything G-ve is increasing.

Good Luck♥