

What Should You Know About Biofilms?

A Microbiology Perspective – Part I

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The association between human disease and microbial contaminants in water sources has been documented since the 1850s,¹ and Snow's groundbreaking "Broad Street Pump" investigation into a London cholera outbreak is credited with launching the scientific discipline of water microbiology. Over the years, much has been learned about the types, characteristics, and cross-infection potential of waterborne bacteria, fungi, parasites, and viruses (Table 1). Yet even with established public health regulations for treating public water supplies, waterborne disease outbreaks continue to be reported from the microbial colonization of drinking water; recreational water; hospital water; and water from health care-associated equipment, including dental unit water lines (DUWLs) and handpieces.

With regard to dentistry, the documentation of high bacteria concentrations in coolant water from high-speed handpieces was first published in 1963.² Later studies revealed that the long lengths of hollow bore tubing that deliver compressed air to power handpieces and water to cool them provide an optimal environment for stagnation and microbial accumulation. Water flowing through untreated dental units can rapidly colonize in DUWLs, leading to high bacterial concentrations — ranging from thousands to hundreds of thousands of colony-forming units per milliliter of water (cfu/mL). (The EPA standard for potable water is just 500 cfu/mL or less of non-coliform bacteria.)

Table 1. Representative Waterborne Disease Agents

Bacterial	Parasitic
<i>Escherichia coli</i>	<i>Cryptosporidium</i>
<i>Vibrio cholera</i>	<i>Giardia</i>
<i>Salmonella typhi</i>	<i>Schistosoma</i>
<i>Pseudomonas sp.</i>	
<i>Shigella sp.</i>	Viral
<i>Legionella pneumophila</i>	<i>Hepatitis A virus</i>
<i>Non-tuberculous Mycobacterium sp.</i>	<i>Noroviruses</i>

Readers of this article have attended multiple seminars and webinars discussing dental water infection control protocols and products to help them provide potable water to patients. As a change of pace from those sessions, this two-part series will use a microbiology perspective to specifically focus on biofilms and the infectious disease challenges they present in dental waterlines and other water sources. This article discusses biofilms and the mechanisms and characteristics of their formation and maturation. The second article in the series will explore the potential health problems microbes in biofilms present for both health care providers and their patients.

Please note: These discussions are brief topic overviews only. See the references for in-depth examinations of published research and waterborne infection outbreaks.

What Are Biofilms?

The overwhelming majority of microorganisms in nature exist and thrive primarily by attaching to and growing on diverse living and environmental surfaces.³ In contrast to early descriptions of biofilms as formless, unstructured groups of random microbial forms, biofilms are highly complex in their composition and architecture. Today, a commonly used definition is:

*"... A microbiologically derived sessile community characterized by cells that are irreversibly attached to a substratum or interface to each other, are embedded in a matrix of extracellular polymeric substances that they have produced, and exhibit an altered phenotype with respect to growth rate gene expression."*⁴

Naturally occurring biofilms are heterogeneous in their composition and structures. For example, aquatic/marine environments and municipal water systems can contain fungi, algae, protozoa, larger microscopic worms, and numerous types of bacteria. Because the majority of microbes can exist in these biofilms, it's not an overstatement

to say we live in world of biofilms. The adaptability and resilience of these microbial accumulations in natural and industrial (e.g., health care) systems allows for the development of group survival strategies. In hospital systems, for example, slower-growing microbes in biofilms can exhibit transcribed genetic modifications that can protect them from mechanical removal and antibacterial chemicals, such as antibiotics and disinfectants.

Biofilm Development

Thick biofilms developing in untreated DUWLs within a week initially led to the question of how that could happen when the facility was supplied with potable municipal water. Multiple structural, water flow, and other factors were shown to be responsible; Table 2 lists and briefly describes them.^{5,9}

Another question is how do waterborne organisms accomplish the interactions required to form biofilms? Establishment is commonly considered to occur in four main stages (Figure 1)⁵:

- 1. Bacterial attachment to a surface
- 2. Microcolony formation
- 3. Biofilm maturation
- 4. Detachment and dispersal of bacteria

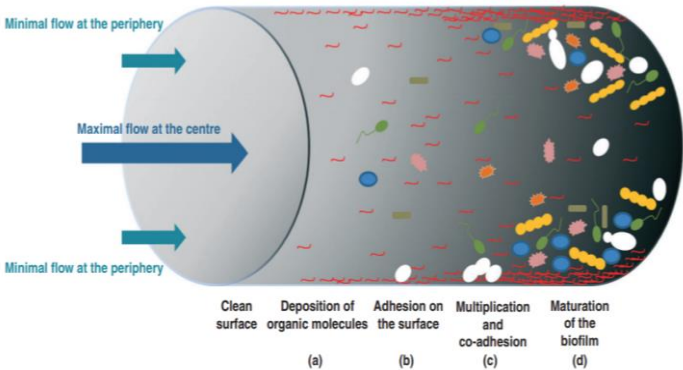


Figure 1. Biofilm Formation in Dental Waterlines⁵

Table 2: Representative Factors Associated With DUWL That Promote Biofilm Growth

- | | |
|----|---|
| 1. | Small cylinder diameter and narrow lumens |
| 2. | System design: dead legs and control blocks |
| 3. | High surface-to-water volume ratio |
| 4. | Tubing materials that are conducive to microbial attachment |
| 5. | Very little water flow at hydrodynamic boundary |
| 6. | Low volume of water used during patient treatment |
| 7. | Water warms to room temperature |
| 8. | Intermittent usage of DUWL |

The last stage can facilitate the attachment and colonization of new areas. Figure 2 illustrates these steps through a scanning electron microscopic guide using dental waterlines. Free-floating (i.e., planktonic) bacteria initially attached themselves to a solid surface and established “pioneer” colonies (Figure 2a).

As microbial growth continues and the layer thickens, an oligosaccharide matrix (called a “glycocalyx”) is synthesized by some members of the resident microflora (Figure 2b). This slime layer provides a firm structural framework that protects organisms within the biofilm from desiccation, exposure to chemicals, and destruction from other microbes. Shielded microorganisms continue to replicate and develop species-specific microcolonies within the biofilm mass. They communicate with each other via chemical messages as they generate additional biofilm layers. Polysaccharide slime water channels within the biofilm offer protection and carry nutrients and oxygen to the growing cells. During the detachment and dispersal stages, the physics of laminar flow of water passing through DUWLs are such that the maximum water flow rate is observed in the center of the lumen, with minimal flow at the periphery. This encourages the initial deposition and adhesion of microorganisms on the tubing surface.^{6,7} As the mass grows and thickens, it eventually comes into contact with the more rapidly flowing water. Eventually, clumps of material are dislodged and exit in water as solid material or as microscopic planktonic organisms (Figures 2c and 2d).

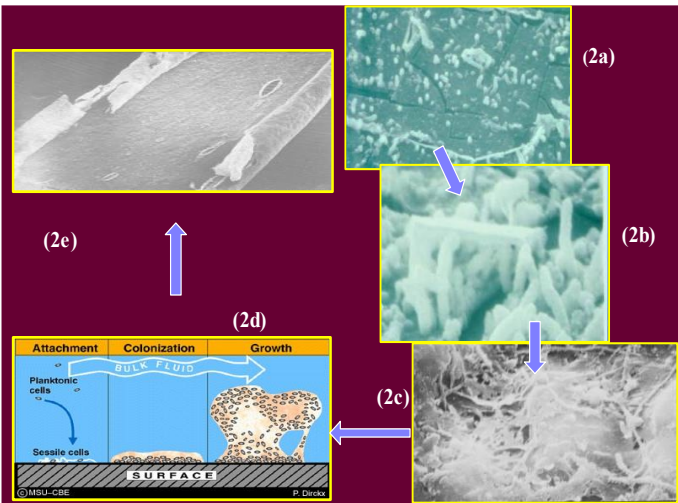
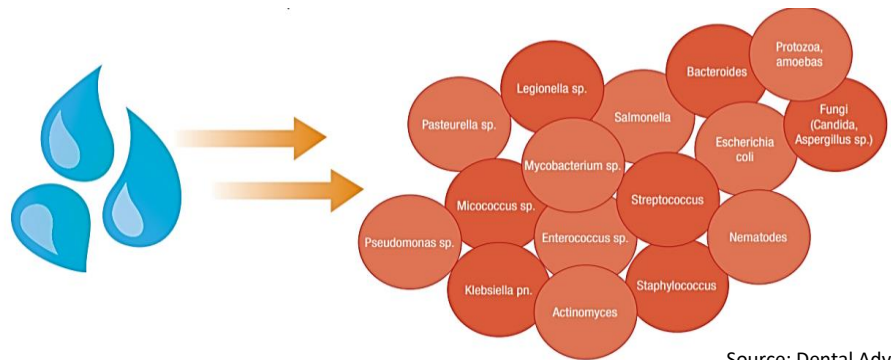


Figure 2. Development of Biofilm in Dental Waterlines

Multiple types of microorganisms have been identified in sampled water systems (Figure 3).⁸ Most of the isolated microbes typically originate from the environment and municipal water sources and don't usually pose a high infection risk for healthy people with competent immune defenses.

However, an increasingly larger percentage of the population (and dental patients) are living with conditions that weaken and compromise the host immune system. When these immune systems are exposed to colonized water, the so-called "non-pathogens" can overcome inadequate host defenses, act as opportunistic pathogens, and cause clinical infections.



Source: Dental Advisor

Figure 3. Representative Microbes Isolated from Dental Unit Waterlines.

In part 2 of the series, we'll discuss the potential risks these organisms present to health care professionals and their patients.

We want to thank Dr. Molinari for answering our questions, and we invite you to evaluate our wide range of products designed to protect clinicians, patients, and the practice during dental procedures.

References

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