THE FLOSSOPHY OF ORAL HYGIENE:
THE RELATIONSHIP BETWEEN PERIODONTAL DISEASE
AND CARDIOVASCULAR DISEASE

By

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To my Mom and Dad,
Thank you for pushing me this far.
I can’t wait to see what our next adventure holds.
Love, Tutie Pie
The Flossophy of Oral Hygiene: The Relationship Between Periodontal Disease and Cardiovascular Disease

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Abstract
It has been estimated that approximately half of the US population has some level of periodontal disease (Paul, 2015). The following paper analyzes four different studies that support my hypothesis which states that a relationship does exist between periodontal disease and cardiovascular disease. For example, in rabbits, it was observed that periodontal inflammation stimulated by *P. gingivalis* dramatically increased lipid deposition in the rabbit’s arteries (Jain, A. et al., 2003). Moreover, it was demonstrated that statins have a pleiotropic effect on arterial and non-arterial inflammation. Along with my literature research, I educated first-graders about the importance of oral hygiene. The first part of the lesson included a PowerPoint presentation about a superhero tooth stopping a villain from causing cavities. Following the presentation, the students participated in an activity where they sorted food and activities into different categories based on healthy and unhealthy habits. Overall, this study confirms a relationship between periodontitis and cardiovascular disease in US adults; and encourages public health educational programs to educate cavity prevention starting at an early age.

Introduction
Periodontal disease is highly prevalent among adults in the United States and has become a critical public health concern. The epidemic of periodontal disease in the United States is being measured through the National Health and Nutrition Examination Survey (NHANES). From 2009-2014, the NHANES implemented a Full Mouth Periodontal Examination (FMPE) protocol that recorded six different probing measurements collected from each tooth (Eke, et al., 2015). This protocol allowed NHANES to create a standard definition of periodontitis, and minimized misdiagnosis in the dental office. Using this new protocol, it was reported that in 2009-2012, 46% of US adults had periodontal disease based on a population of 64.7 million people. Based on this population, 29.7 million adults were diagnosed with periodontitis, and 5.8 million adults (8.9%) were suffering from severe periodontitis (Eke, et al., 2015). Based on these reported statistics, it is evident that there is a critical oral health epidemic in the US.

According to the 2017 update reported by the American Heart Association (AHA), there is an estimated 92.1 million adults living in the US who have at least one type of cardiovascular disease (CVD). The AHA predicts that by 2030, 43.9% of adults in the US will suffer from at least one type of CVD (AHA, et al. 2017).

With the rise of periodontal disease and cardiovascular disease in the US it is imperative that health care professionals begin to design treatments that can affect both disorders; and implement educational outreach programs to educate the public about these diseases. In the following report, I review multiple research studies that have found periodontal disease, if left untreated, to be a significant risk factor for cardiovascular disease. Based on the data provided, it is important for the public to understand the role of oral infections in CVD as well as the actions that oral bacteria plays in local and systemic inflammation.

Cardiovascular System
The cardiovascular system, more commonly known as the circulatory system, is composed of three basic components: the heart, blood vessels and blood. In general terms, the cardiovascular system provides oxygen and nutrients to different
parts of the body that are essential for life, while also removing waste and toxins from the body.

**Anatomy of the Heart**
The heart is a hollow, muscular organ about the size of a clenched fist (Cohen). It lays in the middle of one’s chest between the sternum in the front and vertebrae in the back.

**The Heart Muscle**

![Figure 1. Layers of the Heart. Illustration of the three layers of the heart and their respective locations.](image)

The heart wall is made up of three layers: the pericardium (epicardium), myocardium, and endocardium (Figure 1) (Cohen). The pericardium (including the epicardium) is a double-walled membranous sac that encloses the heart. In other words, the pericardium is the sac that the heart sits in. The pericardium has three functions:

- It helps keep the heart where it belongs (middle of the chest).
- It serves as a layer of protection because by surrounding the heart, it makes it harder for pathogens to get inside and cause infection.
- The pericardial sac has stretch and pain sensors that can act as extra protective mechanisms.

The epicardium is the inner portion of the pericardium that is against the wall of the heart (Cohen). The visceral and parietal pericardia are separated by a fluid-containing space known as the pericardial cavity. The pericardial cavity is filled with pericardial fluid that allows the heart to contract and relax without pain (friction).

The myocardium is the middle and thickest layer of the heart wall. It is composed of cardiac muscle and is affixed to the heart’s fibrous frame that helps the heart open the designated valves (Cohen).

The endocardium is the innermost layer of the heart wall that is made up of a unique type of epithelial tissue that lines the entire circulatory system called endothelium.

**Fibrous Elements in the Heart**
Like muscles, the heart needs something to work against, and these fibrous elements make up these attachments that allow the heart to contract and act as a big muscle. In addition to these attachments, the fibrous elements also make up the heart valves (Figure 2).

There are two types of valves: atrioventricular valves and semilunar valves. Atrioventricular valves are positioned between the atrium and its respective ventricle. The right AV valve is also known as the tricuspid valve; and the left AV valve is often called the bicuspid, or mitral valve (Cohen). The blood in the right ventricle is pushed out to the pulmonary artery, and the blood in the left ventricle is pushed out into the aorta. Therefore, the semilunar valves are called pulmonary and aortic valves respectively.

![Figure 2. Valves of the Heart. Illustration of the atrioventricular and semilunar valves in the heart.](image)
The heart can be portrayed as a “dual-pump”, meaning that the right and left sides of the heart have different functions (Cohen). The right side of the heart pumps blood from the heart to the lungs. This special type of circulation is called pulmonary circulation because it pumps deoxygenated blood from the heart to the lungs where it receives oxygen. In contrast, the left side of the heart pumps blood from the heart to the rest of the body; therefore, the left side of the heart plays an important role in systemic circulation (Figure 3).

**Circuit of Blood Flow**
Following a single drop of blood coming in from the body it enters the right atrium first through the tricuspid valve, and then into the right ventricle where it is pushed through the pulmonary valve and into the pulmonary artery away from the heart and into the lungs. The pulmonary artery transports the deoxygenated blood to the lungs where carbon dioxide is released and oxygen is received during respiration (Cohen). The now oxygenated blood returns through the pulmonary vein and is now collected in the left atrium. When pressure is high enough the blood will push through the mitral valve into the left ventricle. The left ventricle carries oxygenated blood up through the atrial valve and out into the systemic arteries, to the capillaries in the tissues where gas and nutrients exchange occurs, and then transports the now deoxygenated blood through a system of veins and back into the right atrium of the heart ready to start the circuit all over again (Cohen).

**Cardiovascular Disease**
Also referred to as coronary artery disease, or CAD, is a pathophysiologic defect that weakens the pumping ability of the heart; as a result, depriving the heart muscle of oxygen and nutrients (Cohen). In a person suffering from coronary artery disease, the severe plaque buildup in their arteries causes a decrease of blood flow. Because blood is unable to flow normally through the vessels, the tissue behind the plaque begins to grow ischemic. If the blockage is too severe, we can begin to see cell death. This is referred to as a myocardial infarction, more commonly known as a heart attack (Cohen).

**Risk Factors**
Risk factors come in two categories:
- Conventional vs. Non-traditional
- Modifiable vs. Non-modifiable

Conventional, or major, risk factors for CAD that are non-modifiable include advanced age, male gender or women after menopause, and genetics. When measuring age as a risk factor, physicians include men over the age of 45, and women over the age of 55. Women are protected longer because of the levels of estrogen they produce until menopause.
Modifiable major risk factors are risk factors that we can change if need be. These can include, but are not limited to:
- Dyslipidemia
- Hypertension
- Cigarette smoking
- Diabetes and insulin resistance
- Obesity
- Non-active lifestyle
- Atherogenic diet

Non-traditional factors are risk factors that are being discovered with today’s technology and include increased serum markers for inflammation, and infection such as C-rp, fibrinogen and inflammatory cytokines (Cohen).

Atherosclerosis
Consuming an atherogenic diet is considered a modifiable major risk factor for coronary artery disease. An atherogenic diet is a diet that is high in fat, cholesterol and salt. Dieticians explain that people who consume fast-food daily are on a high-fat diet. Fast-food diets promote atheromas, which are plaques lodged inside an artery that has become inflamed. Atheromas are the hallmark of a type of coronary artery disease classified as atherosclerosis.

![Blood Vessel](image)

**Normal and Partly Blocked Blood Vessel**

*Figure 4. Progression of atherosclerosis.*

In simple terms, atherosclerosis occurs when plaque clogs the arteries (Figure 4). As the plaque continue to increase in size, it can limit the blood flow, and the heart will not receive enough oxygen to function properly. When plaque occurs, there is a possibility that the fatty deposit may break off and be carried through the blood stream until it gets stuck; or a blood clot may form on the surface of the plaque (Cohen). If either of these two things transpire, the artery may become completely occluded and no blood will be flowing to the brain and heart. If there is complete blockage, a stroke or heart attack will occur.

**Infection**
By increasing white blood counts during an infection, specifically increasing the number of macrophages recruited, the risk of developing atherosclerosis is also increased. The relationship between infections in the mouth and inflammation in the artery walls is the main discussion of this paper, and includes reliable data and correlation to support that periodontal infection is a non-traditional risk factor for coronary artery disease.

**Dental Anatomy: Introduction**
Dental anatomy is dedicated to the study of the human tooth and its surrounding structures. Dental anatomy covers the development, appearance and classification of teeth. In general, humans have 20 deciduous “baby” teeth and 32 permanent teeth, this is including the third molars more commonly known as the “wisdom” teeth. Among these 32 teeth, 16 are in the maxilla (upper jaw), and the other 16 are found in the mandible (lower jaw). Oral hygiene plays a critical part in the development and appearance of the teeth. By practicing proper oral hygiene, one can prevent dental problems such as cavities, gingivitis, periodontal disease and bad breath; and ensure that their teeth develop properly and stay healthy for a lifetime.

**Anatomy of the Tooth**
The tooth is divided into two sections: crown and root. The term “crown” can refer to either the anatomical crown or the clinical crown. The anatomical crown refers to the portion of the tooth that is covered by enamel. It is the area of the tooth that is above the neck of the tooth. The clinical crown, is the portion of the tooth that is visible in the mouth. In contrast to the crown, the tooth is also composed of an anatomical and clinical root. The anatomical root is the portion of
the tooth that is covered in cementum; whereas, the clinical root is the portion of the tooth that is not visible in the mouth. In addition to the crown and root of the tooth there are four major tissues that the teeth are made of: enamel, dentin, cementum and dental pulp (Figure 5).

**Figure 5.** Anatomy of the tooth and its supporting structures.

**Surrounding Structures**
Along with the tooth, there are many surrounding tissues that support the teeth. The alveolar process is the thick part of the maxilla and mandible that forms a tooth socket to hold and support the teeth. The alveolar process ultimately forms the dental arches (Listgarten, 2016). These sockets are crucial to the tooth’s survival. The alveolar bone is a part of the alveolar process that lines the sockets and where the roots of the teeth attach. In addition to the alveolar bone, the periodontal ligament is another supporting tooth structure that connects the cementum covering the root of the tooth to the alveolar bone (Listgarten, 2016).

In addition to the tooth structures that are responsible for holding the teeth in place, the gingiva is a mucous-membrane tissue that is also involved in the protection of the teeth. Gingiva, more commonly known as gums, immediately surround the teeth. The gingiva is attached to the tooth forming a seal that protects the rest of the tooth and supporting structures from any invading bacteria (Listgarten, 2016).

**Gum Disease**
Periodontal disease, also known as gum disease, can range from simple gum inflammation to more serious conditions that can result in the loss of bone and other supporting structures. The human mouth houses a wide variety of bacteria. These bacteria can begin to build up on the teeth and create plaque. If not taken care of, this plaque can create an inviting niche for periodontal bacteria to live in and consequently cause inflammation of the gingiva. There are two types of gum disease: gingivitis and periodontitis.

**Gingivitis**
Gingivitis is the most common form of periodontal disease. Someone affected with gingivitis clinically has red, swollen gums that bleed easily during flossing or brushing (American Academy of Periodontology). However, it is important to note that there is usually no pain associated with gingivitis. If caught in time, gingivitis can be treated easily with regular visits to the dentist and maintaining a good oral health care routine at home.

**Figure 6.** Diagnostic photo of gingivitis. Please take note of the swollen, red gingiva.

**Periodontitis**
If left untreated gingivitis can advance to periodontitis. The plaque growing on the tooth can continue to build up, and over time it spreads below the gum line. Now the periodontal bacteria can release toxins underneath the gum line that incites a chronic inflammatory response (American Academy of Periodontology). This
ongoing inflammation ultimately causes the supporting tissues and bone of the teeth to be broken down. Along with the destruction of supporting tooth structure, periodontitis is also characterized by recessive gum lines, periodontal pockets, and teeth can become loose and fall out.

**Risk Factors**
Everyone is susceptible to gum disease if they do not maintain a proper oral health care routine. However, there are certain risk factors that can help expedite the process of tooth destruction such as smoking, diabetes, medications and hormonal changes in girls/women (NIH, 2013).

**Figure 7.** Diagnostic photo of periodontitis. Take note of the swollen gums that are beginning to recede revealing the root of the tooth.

**Treatment**
The main goal of treatment for gum disease is to control the inflammation and infection. The standard care for periodontal disease is a scaling and root planing, SRP. Usually the dental hygienist removes the built-up plaque through a deep-cleaning that includes scraping off the tartar above and below the gum line (scaling), and eradicating any rough spots on the root of the tooth where bacteria congregate (root planing). Along with SRPs the dentist might prescribe medication to help with inflammation, and create a proper oral health care routine for the patient to follow at home.

**The Oral Microbiome**
The oral microbiome plays a significant role in balancing health and disease in a person. Within the oral cavity, the microorganisms that live there have been linked to many oral and systemic pathologies. In the following sections, I will be addressing both the local and systemic pathologies that oral microorganisms have been linked with.

The oral microbiome consists of bacteria, archaea, fungi, protozoa and viruses that contribute to the health of the host through various relationships.

**Bacteria in the Mouth**
The major bacteria phyla living in the oral cavity of an adult include Firmicutes, Bacteroidetes, Proteobacteria, Actinobacteria, and Fusobacteria. The most prevalent genus within the oral microbiome is *Streptococcus* (Sampaio-Maia, 2016) The Human Microbiome Project (HMP) assessed the bacteriome from the cheek, hard palate, keratinized gingiva, palatine tonsils, saliva, subgingival and supragingival plaque, throat and tongue in over 200 healthy individuals (Sampaio-Maia, 2016). The results of this study are depicted
in figure 8. The genera in bold are presented in more than 75% samples that had a population greater than 10%. The non-bolded genera were greater than 1% abundance found in more than 80% of the samples.

In the oral cavity, bacteria populations exist as biofilms. Biofilms are three-dimensional structured communities that are highly organized and attached to a surface (Baltrus, Lecture 34). Within biofilms, bacteria interact by quorum sensing. Quorum sensing allows individual bacteria to act as a multicellular organism. Through the production of chemical signals, bacteria can monitor population density and regulate gene expression (Baltrus, Lecture 31). This is significant because oral biofilms are present on all oral surfaces, but the biofilms on teeth, also known as dental plaque, can cause decay. Dental plaque is a mass of bacteria that grows on the surfaces in the mouth that can eventually form tartar on or around the teeth. It begins with the attachment of early bacteria colonizers to tooth enamel (Sampaio-Maia, 2016). With the attachment of early bacteria colonizers, microorganisms that are unable to bind directly to the tooth can now bind and create biofilms on the enamel. If left untreated, the plaque continues to build up and can cause tooth decay and eventually chronic inflammation of the gingiva and bone loss.

The Oral Microbiome and Periodontal Disease
The oral microbiome plays a significant role in maintaining health in the oral cavity, and fighting off certain diseases. These oral diseases can occur when microbes become imbalanced. In other words, when microbial populations are reduced in the oral cavity, or relocated to a different area where they do not belong. Periodontal disease occurs when oral bacteria becomes abundant in oral tissues that results in inflammation and destruction of teeth and their supporting structures.

Periodontal diseases include, but are not limited to, gingivitis and periodontitis. Periodontitis is a very aggressive form of gingivitis that causes progressive destruction of periodontal tissues, like the alveolar bone, resulting in the formation of periodontal pockets (Figure 9).

The process begins with local inflammation targeted around the soft tissues of the teeth, more commonly known as the gums. This inflammation, also known as gingivitis, is caused by biofilms (plaque) that forms on the tooth surfaces at the gum line. If left untreated it will progress to periodontitis which will then lead to irreversible destruction of important tooth structures as stated above.

Figure 9. Periodontal pockets in healthy and diseased individuals.

It is commonly accepted that periodontitis is caused by the interaction of soft subgingival biofilms with a down regulated host response in the periodontal tissues (Sampaio-Maia, 2016). However, the precise role of these biofilms, dental plaque, in periodontal disease is still unclear. Several studies have found many species of oral bacteria in atherosclerotic plaque including Streptococcus spp., Ceillonella spp., P. gingivalis, A. actinomycetemcomi-tans, T. denticola, F. nucleatum, T. forsythia, and Neisseria spp (Sampaio-Maia, 2016). A recent study observed 84 different bacterial taxa that were found in the lining of the artery walls of patients with ASVD and periodontal disease, while only 18 different taxa were found in the vascular walls of patients with only vascular disease (Armingohar, et al., 2014). These results propose the idea that the oral cavity may be a potential source for the dispersal of bacteria into vascular tissues. Below are several hypotheses that try to explain the role of microbes in oral pathology.
Specific Plaque Hypothesis
The specific plaque hypothesis states that a few species of Gram-negative anaerobes are responsible for periodontal disease (Loesche, 1992; Loesche & Grossman, 2001). In 1998, the microbial complexes most commonly associated with periodontal disease were the orange micros, and *F. nucleatum*, and the red complex, consisting of *P. gingivalis*, *Tannerella forsythis*, and *Treponema denticola* (Sampaio-Maia, 2016). More recently, specific pathogens such as *P. gingivalis* and *F. nucleatum* have been associated with periodontitis and its complex relationship with systemic disease (Sampaio-Maia, 2016).

Non-Specific Plaque Hypothesis
The non-specific plaque hypothesis states that all the microorganisms living in the oral cavity contain the virulence factors necessary to cause inflammation and decay of periodontal structures (Loesche & Grossman, 2001).

Ecological Plaque Hypothesis
The ecological plaque hypothesis states that the environmental conditions inside the periodontal pockets is what causes the expression of virulence factors in the bacteria. These factors are already present, but in small quantities (Marsh, 2003). This hypothesis helps explain why bacteria can cause disease in certain locations, but not in others.

Regardless of the mechanism adopted to cause periodontal disease, all three hypotheses show that disease is caused by a shift in the composition of the oral microbiome from a clinically healthy gingiva, into a diseased state.

The Oral Microbiome and Systemic Pathology
Oral microorganisms can travel through oral mucous membranes and periodontal pockets and eventually maneuver to different locations of the body ultimately causing systemic diseases. Even though the high prevalence of periodontitis and cardiovascular disease, CVD, makes it a challenge to evaluate the relationship between these conditions; there is reliable and reasonably strong evidence linking periodontitis to CVD. This relationship is supported by animal and clinical studies that will be discussed below.

Cardiovascular Disease
Numerous studies have proposed that people with periodontal disease have a higher risk of developing cardiovascular disease. Researchers have proposed a variety of mechanisms that link oral infections to cardiovascular disease. It is universally accepted that inflammation plays a

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**Figure 10.** Radiographs of the jaws of experimental models (left picture) and the control animals (right picture). The radiograph on the left shows evident bone loss in the second premolar area (red arrows).
significant role in both oral infections and CVD. Inflammation in the oral cavity is stimulated by the bacteria in dental plaque that forms on the tooth surfaces and in the gingival margin (gum line). Likewise, vascular inflammation is caused by many risk factors such as hyperlipidemia, hypertension, and smoking. One generally accepted pathophysiological mechanism that ties the oral microbiome with systemic diseases is based on the “colonization” of atherosclerotic plaque with oral microorganisms that causes systemic inflammation (Kholy et al., 2015). This hypothesis, that local inflammation can cause an inflammatory response systemically and ultimately lead to atherosclerosis is discussed below.

**Periodontitis Increases the Risk for CVD**

One critical trait that researchers have been trying to understand is this idea that focal infections can cause systemic inflammation. Most of the studies done to investigate the mechanisms and biological possibility of bacteria found in the mouth to be causing a systemic inflammation in the vasculature is performed in animals.

In the following study, periodontal inflammation in rabbits stimulated by *P. gingivalis* was seen to dramatically increase lipid deposition in the rabbit’s arteries (Jain, A. et al., 2003). Jain Ashish and fellow researchers evaluated the effects of periodontitis on cardiovascular outcomes of a high-fat diet in a rabbit model. In their study, New Zealand White rabbits were maintained on a 13-week high-fat diet consisting of 0.5% fat to encourage the buildup of lipid deposits in the aorta. The experimental group received silk ligatures around their mandibular premolars followed by a topical application of *P. gingivalis* to promote periodontitis in the animal model. The results of the experiment are as follows. Table 2 shows the analysis of bone levels in the test group and the control group. The average crestal and proximal bone loss in the rabbits induced with periodontitis was significantly greater than in the control group with the exceptions of rabbit 54 and 55. To support this data, the researchers obtained radiographs to observe the difference of bone loss in both groups (Figure 10).

Moreover, along with recorded bone loss, figure 11 shows the histologic sections of the rabbits

---

**Table 2.** Changes in the periodontal supporting bone after 14 weeks in the periodontitis and control groups.

<table>
<thead>
<tr>
<th>Group and rabbit</th>
<th>Mean bone loss (mm)*</th>
<th>% Tooth in bone</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Crestal</td>
<td>Proximal</td>
</tr>
<tr>
<td>Periodontitis</td>
<td>54</td>
<td>4.41</td>
</tr>
<tr>
<td></td>
<td>55</td>
<td>3.76</td>
</tr>
<tr>
<td></td>
<td>56</td>
<td>10.24</td>
</tr>
<tr>
<td></td>
<td>57</td>
<td>7.67</td>
</tr>
<tr>
<td></td>
<td>58</td>
<td>5.16</td>
</tr>
<tr>
<td>Mean ± SEM</td>
<td>6.24 ± 2.67*</td>
<td>4.9 ± 1.46*</td>
</tr>
<tr>
<td><strong>Control</strong></td>
<td>59</td>
<td>4.33</td>
</tr>
<tr>
<td></td>
<td>60</td>
<td>3.91</td>
</tr>
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<td></td>
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<td></td>
<td>62</td>
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<td>63</td>
<td>2.75</td>
</tr>
<tr>
<td></td>
<td>64</td>
<td>4.58</td>
</tr>
<tr>
<td>Mean ± SEM</td>
<td>3.63 ± 0.81</td>
<td>3.31 ± 0.53</td>
</tr>
</tbody>
</table>

*Expressed as the distance between a fixed reference point (tooth crown tip) and the bone crest.

Significantly different from control group at a *P* of <0.05.

Significantly different from control group at a *P* of <0.005.

---

**Table 2.** Analysis of changes in supporting bone levels of rabbit models in the periodontitis and control groups.
after 13 weeks. In the group of rabbits with periodontal disease, their tissue assessment of bone resorption revealed increased cellular infiltration, alveolar bone resorption, a loss of connective tissue organization (Jain, A. et al. 2003). This is significant because these three characteristics are also key traits of chronic periodontitis in humans. Finally, figure 12 displays the amount of lipid deposition in the aortas of rabbits with and without periodontal disease. The top panel reveals that periodontitis and associated bone loss were related to widespread aortic lipid deposition. In contrast, the bottom panel depicts that the control rabbit had minimal lipid deposition.

Ashish and fellow researchers concluded that in the rabbit high-fat diet model, animal models with experimentally induced periodontitis developed extensive lipid deposits in their aortas compared to their healthy counterparts. This evidence suggests that remote inflammation in the oral cavity can have direct effects on the innate immune system response in systemic organs; as a result, increasing the risk for inflammation and tissue injury.

**Invasion Methods of Oral Bacteria into the Vasculature**

Once someone has periodontal disease, there is evidence for two modes of invasion into the cardiovascular tissue by periodontal pathogens: bacteremia and phagocyte-mediated bacterial translocation (Carrión et al., 2012; Kinane, Riggio, Waalker, MacKenzie & Shearer, 2005).

**Bacteremia**

Periodontal pockets are a portal for bacterial entry into systemic circulation. This results in bacteremia (Kholy et al., 2015). As the bacteria invade the endothelial layer, they produce pro-inflammatory cytokines such as monocyte chemoattractant protein 1, MCP-1.

Chemokines are pro-inflammatory cytokines that serve to attract and activate leukocytes. Moreover, they are connected to the pathogenesis of atherosclerosis, and the expression of the CC chemokine monocyte chemoattractant protein -1 (MCP-1) is upregulated in human atherosclerotic plaques. Boring and his fellow scientists investigated whether MCP-1 is associated with the development of atherosclerosis by producing mice that lack the receptor for MCP-1, CCR2, and crossed them with apolipoprotein E-null mice (apoE/−) who have severe atherosclerosis. They discovered that the selective absence of CCR2 decreases lesion formation in apoE/− mice.

Figure 13 illustrates macrophage infiltration of the aortic sinus in apoE/− mice fed a high-fat diet for five weeks. Sections from the aortic sinus of the mice were stained red for macrophages with
MOMA-2. Macrophages were abundant of CCR2<sup>+/+</sup>, apoE<sup>−/−</sup> mice and comprised most of the cells in the aortic lesion (Fig. 13a). In contrast, fewer macrophages were visible in the aortic lesions of CCR2<sup>−/−</sup>, apoE<sup>−/−</sup> mice (Fig. 13b). After staining, the researchers calculated the mean macrophage area and concluded that there was significantly less MOMA-2 positive staining in CCR2<sup>−/−</sup> mice than in CCR2<sup>+/+</sup> mice (Fig. 13c). The abundance of macrophages seen in CCR2<sup>+/+</sup> mice suggests that activation of CCR2 is important in macrophage recruitment into the vessel wall, which is the earliest identifiable sign of atherosclerosis (Boring, 1998).

Along with the data that suggests that CCR2<sup>+/+</sup> mice have increased macrophage recruitment, Boring also discovered by staining with Oil Red O that CCR2<sup>−/−</sup> mice had a significant decrease in lesion area compared to mice who do express the CCR2 gene. This data indicates that the reduced recruitment of macrophages observed at five weeks had smaller lesion sizes the rest of the trial (weeks 9-13). In summary, the above study identified CCR2 as a pre-determined genetic trait for atherosclerosis, and provided strong evidence for a direct effect of MCP-1 in monocyte/macrophage recruitment and atherogenesis (Boring, 1998).

**Phagocyte-Mediated Bacterial Translocation**

On the other hand, because of this increased population of bacteria inside these pockets, there is also an increase in the number of phagocytic cells moving to the gingival sulcus. These phagocytic cells can transport bacteria from the gingival sulcus to remote sites such as vascular tissues.

A recent study suggests that dendritic cells phagocytose periodontal pathogens from diseased periodontal pockets and transports them through the bloodstream into vascular sites (Carrion, et al., 2012). The study followed healthy and periodontitis patients with or without atherosclerotic vascular disease, ASVD. In the current study, they show that blood myeloid dendritic cells, mDCs, provide periodontal microbes with a protective niche and mode of
transportation. In return, *P. gingivalis* initiates differentiation of mDCs into a phenotype that promotes the growth of atheromas.

The researchers observed that blood myeloid dendritic cells carry known periodontal pathogens such as *P. gingivalis*, and that the number of dendritic carrier cells increased in patients who suffered from both periodontal disease and ASVD. The results show that the percentages of blood mDCs were the lowest in the healthy control group, and the highest in patients who suffer from coronary artery disease and chronic periodontitis, CP (Figure 14).

The next part of their study was determining whether the blood mDCs housed periodontal microbes. Blood mDCs were isolated from CP and ASVD/CP patients and they were analyzed using 16s rDNA sequencing, immunofluorescence, and viable bacterial cultures (Carrion, et al. 2012). Seventy-two percent of CP individuals who were exposed to the microbe were found to have *P. gingivalis* 16S rDNA in their blood mDCs; whereas no mDCs in the control group were positive for this trait. Researchers also looked for a correlation that existed between elevated peripheral blood DC counts with the presence of a blood-borne infection. To do so, they had the CP patients undergo SRP treatment, the standard care for patients with chronic periodontitis. Based on previous studies, it is known that this kind of mechanical debridement can drive oral bacteria into the blood stream of the patient (Carrion, et al. 2012). After debridement, they observed a significant increase in *P. gingivalis* inside mDCs (Fig. 15C), and a 25% increase in circulating blood mDCs (Fig. 15D).

Moreover, now that they observed that there is a positive correlation between elevated blood DC

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**Figure 14.** Increased blood mDC frequency. (1) Mean numbers of mDCs (2) Percentages of mDCs (3) total numbers of PBMCs in CTP, CP, and ACS/CP groups

**Figure 15.** Increased blood mDCs after local debridement. (C) Significant increase in *P. gingivalis* seen in blood mDCs 24 hours after local debridement. (D) Scattergrams from a flow cytometry analysis of blood mDCs at 0 hours and 24 hours after a local debridement.
counts and blood-borne infection, Carrion and her fellow researchers considered the differentiation of DCs from monocytes that can be enhanced by ligation of DC-SIGN by *P. gingivalis* mfa-1. When in the presence of *P. gingivalis* at a MOI of 0.1, mDC differentiation was increased by 28%; as a result, increasing the growth of the atheroma by 28%. This further supports that mDC differentiation can ultimately convert mDCs into an atherogenic phenotype.

The study concluded that dendritic cells in the blood play a significant role in dispersing oral bacteria to atherosclerotic plaques. In addition, they proposed that these oral organisms provide necessary signals for dendritic cell differentiation and contribute to the growth of the atherosclerotic plaque once they enter the artery wall. Ultimately, this mechanism between oral bacteria and dendritic cells reveals how local infections can increase the risk of cardiovascular disease.

**Treatment for PD and ASVD**

There are many different treatment methods for periodontal disease. They range from scaling and root planing, medication, and change in diet. However, there was not a treatment plan used for both periodontal disease and cardiovascular disease. Previously, in 2013, Subramanian and his fellow researchers began to investigate the role of statins.

**Statin Therapy**

Statins have been found to have positive effects in treating atherosclerotic diseases. They are said to successfully decrease the amount of low-density lipoprotein cholesterol in the blood, and reduce systemic inflammation caused by atherosclerotic plaque (Subramanian, S. et al. 2013).

In a recent study, they demonstrated that using statin therapy not only reduces cardiovascular inflammation, but also reduces periodontal inflammation in the same patients. Subramanian and his fellow researchers used FDG-PET computed tomography (CT) imaging to evaluate an innovative effect of statin treatment on periodontal tissue. They hypothesized that Atorvastatin treatment would not only lower periodontal disease activity, but also atherosclerotic plaque activity; as a result, supporting that there is a relationship between both disease (Subramanian, S. et al. 2013).

In their study, eighty-three adults either with risk factors or with current atherosclerosis were put in a randomized, double blind trial to evaluate the effectiveness of Atorvastatin on arterial inflammation. Subjects were randomized to take a 10-mg Atorvastatin tablet plus an 80-mg
Atorvastatin matching placebo daily; or an 80-mg Atorvastatin tablet plus a 10-mg Atorvastatin matching placebo. Their arterial and periodontal tracer activity was measured along with the amount of periodontal bone loss after twelve weeks (Subramanian, S. et al. 2013).

At first, they evaluated the relationship between periodontal disease and periodontal FDG uptake at baseline. The FDG uptake, also known as TBR, at baseline was compared with CT images that showed alveolar bone resorption. A PET scan uses a radioactive drug, or tracer, to show differences between healthy tissue and diseased tissue. Fluorodeoxyglucose, FDG, is one of the most commonly used tracers. Before the PET scan, a small amount of FDG is injected into the region of interest and the FG uptake, TBR, is measured (ACRN). Diseased tissue normally grows faster than healthy tissue, so it is assumed that the unhealthy tissue will have a higher TBR than its healthy counterpart. Figure 16 confirms that the mean baseline TBR was significantly higher in areas that suffered from alveolar bone loss than regions that did not have any recorded bone loss at all.

When analyzing the relationship between statins and periodontal tissue inflammation they observed a normal distribution of periodontal FDG uptake values in both treatment groups at baseline, week four and week twelve. However, there was a significant decrease in periodontal FDG uptake after twelve weeks in subjects who have been taking high-dose Atorvastatin versus subjects taking low-dose Atorvastatin. In patients who were taking high-dose of Atorvastatin, their mean change TBR was -0.29 +/- 0.85 vs. 0.13 +/- 0.68 seen in patients taking a low-dose (Subramanian, S. et al. 2013). Figure 17 reveals the relationship between atherosclerotic inflammation and periodontal inflammation. They learned that the baseline TBR in periodontal tissue was proportional with baseline FDG uptake in carotid plaque.

In addition, there were changes in periodontal inflammation that were directly related with changes in carotid inflammation seen after twelve weeks of statin therapy. These changes are recorded in figure 18.

All in all, this study supported that twelve weeks of high-dose (80-mg) Atorvastatin therapy drastically lowers periodontal inflammation; and based on the results of the study, they concluded that statins may influence arterial and non-arterial (periodontal) inflammation; and reducing one can lead to the decrease of inflammation in the other (figure 19).
Works Cited


# Oral Hygiene

**OVERVIEW**

The students will explore the importance of oral hygiene. They will learn the importance of teeth, how to maintain healthy teeth, and what happens when teeth are not taken care of. The first part of the lesson will include a PowerPoint presentation about a superhero tooth stopping a villain from causing cavities. Following the presentation, the students will participate in an activity where they sort food and activities into different categories based on healthy and unhealthy habits. At the end of the lesson, I will distribute goodie bags to the students that include a toothbrush and toothpaste.

<table>
<thead>
<tr>
<th>What I want to happen</th>
<th>Reflection</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>OBJECTIVES</strong></td>
<td>The students did understand why brushing their teeth was so important, and they said they would start brushing their teeth more with their new toothbrush.</td>
</tr>
<tr>
<td>The students will investigate the importance of oral hygiene, and apply what they learned to their lifestyles.</td>
<td></td>
</tr>
<tr>
<td><strong>INFORMATION</strong></td>
<td>The students already knew a lot about how to brush their teeth; however, I think they really learned how cavities occur, and how they can prevent them.</td>
</tr>
<tr>
<td>The students will learn how to brush their teeth, what happens if they do not brush their teeth, and how to fight oral bacteria.</td>
<td></td>
</tr>
<tr>
<td><strong>ACTIVITY</strong></td>
<td>The students loved the activity! However, I wish there were more categories because they were picking up the patterns quickly: fruit is good and candy is bad.</td>
</tr>
<tr>
<td>The students will sort different food items onto the felt board. There will be two different categories: good food for your teeth, and bad food for your teeth.</td>
<td></td>
</tr>
<tr>
<td><strong>SUMMARY</strong></td>
<td>It was a successful presentation and activity! It was a rewarding experience, and I hope to continue it into my future dental career.</td>
</tr>
<tr>
<td>After providing each student with their own toothbrush and toothpaste, I hope to instill healthy habits.</td>
<td></td>
</tr>
</tbody>
</table>

## REQUIREMENTS

- Tooth Care Presentation
- Mouth Felt Board
- Cards for Felt Board
- Toothbrushes
- Toothpaste

## NOTES

Remember to keep the information simple. First-graders are very talkative, so make sure I allow time for every first-grader to answer my questions.
APPENDIX B – COMMUNITY OUTREACH PRESENTATION

For my community outreach project, I designed and delivered an oral hygiene presentation to a class of 21 first-grade students. I believe that I had a very successful presentation. I am first going to discuss the PowerPoint, then the activity, and finally an overall summary of what I learned.

I presented a PowerPoint on one of the many adventures of Timmy the Tooth. In this adventure, Timmy the Tooth is fighting Dr. Bach Teria and his army of sugar bugs using the power of oral hygiene. In retrospect, the presentation itself was successful. The information was succinct and the children could understand it. However, I wish I would have allotted more time for the presentation. I underestimated how many questions first-graders have, and how most of the questions are just stories about whatever is on their mind. However, after the first few slides I learned how to quiet them down, or ask them to wait until after the presentation. I was really impressed on how intrigued they were with the characters. After the story, the students continued to talk about Dr. Bach Teria and Timmy the Tooth. I felt like I connected with them well by having a superhero talk to them about oral hygiene, rather than another adult. The PowerPoint I presented is in Appendix C.

After the PowerPoint presentation, I had the students participate in a group activity. In this activity, I explained to them that Dr. Bach Teria had infected another mouth and we had to stop him from destroying the teeth. To do this, they each had to sort a different food item into two categories: good for your teeth or bad for your teeth. This activity went exceptionally well! The students loved it, and everyone participated. I wish I would have had more categories. If I were to redo this activity I would have more categories: good food vs. bad food, and good habits vs. bad habits. This way the students can learn a variety of ways to prevent cavities. Figure 20 and 21 are pictures of students sorting their food items onto the felt board. Figure 22 is a picture of the mouth after all the cards had been sorted out.

All in all, this community outreach project was a very rewarding experience. I had an excellent time traveling back home and having the opportunity to come and present in my old first-grade classroom. It was worthwhile to see the children’s eyes light up when I was telling them about the adventures of Timmy the Tooth, when I told them they get to fight Dr. Bach Teria, and at the very end when I presented them...
with their own oral hygiene powers (toothbrush and toothpaste). By providing these items I hoped to solidify what they learned in class. Growing up in a low-income public school, I remember almost every presenter who took the time out of their lives to come and teach us. I remember what it was like to want to teach and make a difference in children’s lives; and today I finally got to do just that. In the future, I hope to continue educating the public about the importance of oral hygiene, and inspire young students to achieve the lifestyles they desire.
Take note of the reuse of Dr. Bach Teria. The students instantly understood which side was good vs. which side was bad.
Appendix C – Tooth Care Presentation

ADVENTURES OF TIMMY THE TOOTH

TIMMY THE TOOTH IS GOING TO TEACH YOU:

• WHY TEETH ARE IMPORTANT
• WHAT HAPPENS IF YOU DON’T TAKE CARE OF YOUR TEETH
• HOW TO TAKE CARE OF YOUR TEETH

I need help fighting Dr. Bach Terial! Can you help me?
WHY TEETH ARE IMPORTANT

- HELP YOU CHEW
- HELP YOU SPEAK CLEARLY ("TH" SOUND)
- AFTER YOUR BABY TEETH, YOU ONLY HAVE ONE SET OF TEETH!
- GIVE YOUR FACE SHAPE
- KEEP YOUR BODY HEALTHY!

Ahh Timmy! These teeth are mine! No stopping me!

Why do we need to stop Dr. Bach Teria?
You can’t beat me! My evil plan is underway!

**EVIL PLAN: PHASE 1**

if you keep eating that candy you’ll get a CAVITY!

I TOLD YOU SO!

SUGAR + BACTERIA FORMS ACID

ACID + HEALTHY TOOTH FORMS DECAY
EVIL PLAN: PHASE 2

✓ CAUSE TOOTH DECAY
✓ CREATE TOOTH PAIN

My army of sugar bugs will create holes in the children’s teeth for us to live!

Enamel Decay

Dentin Decay
Evil plan complete! Now time to find the next child!

You will never stop me!

Me and my new friends will stop you with the power of oral hygiene!
**How to Brush Your Teeth:**

1. **Place a pea-sized amount of toothpaste on your toothbrush**
2. **Hold your brush at an angle**
3. **Brush all your teeth from the front to the back, and the top and the bottom. Don’t forget your tongue!**
4. **Brush back and forth, round and round.**
5. **Don’t forget to spit and rinse**
Great work team! You helped stop Dr. Bach Teria!
ADVICE FROM TIMMY THE TOOTH

• Make sure you floss!
• Use mouth wash
• Go visit the dentist every 6 months
• Eat healthy foods
• Avoid sugary foods

AND MOST IMPORTANTLY
BRUSH YOUR TEETH TWICE A DAY!