The Inflammatory Origins of Periodontal Disease and Diabetes: A Framework for Understanding Clinical Outcomes

A Peer-Reviewed Publication
Written by Jeffrey A. Sibner, D.M.D.

Abstract
Inflammatory diseases share many common traits, including the ability to produce pro-inflammatory messengers called cytokines that travel throughout the body. Cytokines play an important role in physiologic regulation of many biological activities, but in inflammatory diseases, they are produced at levels that can have pathological consequences. In diabetes, cytokines can cause β-cell death and insulin resistance. In periodontal disease, the same cytokines are responsible for collagen destruction and alveolar bone loss. Because inflammatory diseases share a common biological denominator, they can often influence the progression of other inflammatory diseases.

Educational Objectives
At the conclusion of this course participants will understand:
1. The role of inflammatory cytokines in periodontal bone destruction and insulin resistance in diabetes.
2. The effect of periodontal disease on blood sugar levels.
3. How to advise patients and predict treatment outcomes for peri patients with diabetes.

Author Profile
Jeffrey A. Sibner, D.M.D. is an Adjunct Assistant Professor at the University of Pennsylvania School of Dental Medicine where he teaches a course on periodontal disease and diabetes. He is affiliated with St. Mary Medical Center and teaches a lay course called Diabetes and You through their diabetes education program. Dr. Sibner maintains a private practice in general dentistry in Bucks County, Pennsylvania.

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Abstract
Inflammatory diseases share many common traits, including the ability to produce pro-inflammatory messengers called cytokines that travel throughout the body. Cytokines play an important role in physiologic regulation of many biological activities, but in inflammatory diseases, they are produced at levels that can have pathological consequences. In diabetes, cytokines can cause β-cell death and insulin resistance. In periodontal disease, the same cytokines are responsible for collagen destruction and alveolar bone loss. Because inflammatory diseases share a common biological denominator, they can often influence the progression of other inflammatory diseases.

In diabetes and periodontal disease, three of the more important cytokines are tumor necrosis factor-alpha (TNF-α), Interlukin-1β (IL-1β) and prostaglandin E, (PGE_2). Hyperglycemia also increases the production of Advanced Glycation End Products (AGE) which interact with collagen, blood vessels and immune cells to cause tissue destruction and increase proinflammatory cytokines.

Because these inflammatory mediators affect both periodontal disease and diabetes, when one disease becomes more severe, it tends to make the other disease harder to control. Conversely, reducing the concentration of inflammatory mediators by treating either disease has been shown to affect the progression of the other disease in a positive manner.

Understanding the common origins of diabetes and periodontitis can help clinicians predict when diabetes is most likely to affect periodontal treatment or periodontal therapy may affect hyperglycemia.

Nature is often very thrifty, using similar biochemical systems in different ways throughout the body. Sometimes, identical peptides are used in completely different ways. For instance, serotonin, a neurotransmitter at nerve synapses throughout the body, is used in a completely different pathway in the brain. Other times, different disease processes can manifest with remarkably similar biochemical profiles. Such is the case with a group of inflammatory diseases that includes periodontal disease, diabetes mellitus, rheumatoid arthritis and irritable bowel syndrome.

It’s not surprising that periodontitis is an inflammatory disease – even the name of the disease indicates inflammation – but it is only relatively recently that we have come to appreciated that it is more than just a local disease – it is a systemic inflammatory disease that can influence other pathologies and conditions.

Perhaps it is less obvious that diabetes is also an inflammatory process. How does the loss of β-cells translate to inflammation? The answer is not obvious – glucose is a normal, necessary constituent in our blood and does not cause inflammation, in and of itself. In fact, the answer is as complicated as the disease, in all of its several forms. Understanding the origins and pathways that lead to inflammation in periodontal disease and diabetes can provide valuable guidance in understanding the two diseases and their systemic impact and in predicting when one disease is most likely to affect the other to exert synergistic complications.

Inflammation in Type 1 Diabetes
Type 1 Diabetes mellitus (T1DM) is an autoimmune disease most commonly manifesting during or just before puberty. Only about 15-30% of all Type 1 Diabetes cases occur in adults 20 years or older. In this form of the disease, β-cells in the pancreas are destroyed by the body’s immune system. Because β-cells manufacture insulin, a hormone required for cells to uptake glucose, the system of control necessary to regulate the glucose pathway is destroyed. This creates a situation where cells are literally starving for glucose, even though there is an excess of glucose in the blood supply.

Other systems become dysfunctional as well. Gluconeogenesis, which is the formation of glucose from non-carbohydrate sources, occurs as cells signal that they need glucose. As glycerol, lactate and proteins are consumed in gluconeogenesis, marked weight loss can occur in the undiagnosed diabetic subject. At the same time, blood levels of glucose become high enough to create hyperosmotic conditions. The kidneys respond by allowing glucose and proteins to be cleared in the urine, and so the uncontrolled diabetic develops glycosuria and proteinuria.

While these changes are pathological, they do not explain the many complications of diabetes that commonly occur, and this is because there is another important pathway at work. Glucose is a high-energy, highly reactive molecule. It can bond with a variety of free amino acid groups in a concentration-dependant fashion – the higher the concentration of glucose in the blood system, the more likely glucose will react with its surroundings.

The most commonly recognized glucose reaction is with hemoglobin. In fact, the level of glycated hemoglobin is so directly proportional to the level of glucose in the bloodstream that it is the standard measure of glucose control in a blood test called Hemoglobin A1c (HbA1c).

Other sites for glycation include epithelial cells, collagen, proteins, lipids and even nucleic acids. Together, these fused structures are referred to as Advanced Glycation End Products or AGE. It is this non-enzymatic glycation from which inflammation arises. The first mechanism by which this occurs is a direct pathway. AGEs interact with many types of
cells, such as fibroblasts and endothelial cells, and with tissue constituents, most notably collagen. Any time tissue is damaged, the immune system gears up to repair the resulting lesion. White blood cells infiltrate the affected tissue, enzymes are released that break down damaged tissue, and repair begins. This type of inflammation tends to be local. But with diabetes, two factors makes the inflammation systemic. First, glycation does not occur in just one area, it occurs everywhere, simultaneously. Since there is too much glucose everywhere, there is damaged tissue and inflammation everywhere as well. AGE affects micro- and macrovascular systems, leading to kidney, heart and eye disease. The neurological system is also affected, leading to a variety of ailments such as numbness or pain in the legs and feet, sexual dysfunction, and digestive problems. AGE-mediated changes to elastin can result in problems from wrinkles to hammertoes.

Perhaps as important is the messenger effect. When infection or tissue damage occurs, PMNs and macrophages are called to the scene. Recruitment takes place via messengers called cytokines. We will examine the “big 3” cytokines involved with bone loss later. For now, it is important to understand that cytokines not only recruit white blood cells locally, but they engage the entire inflammatory system, alerting cells throughout the body that there is something going on, and they should be ready.

Advanced Glycation End Products also stimulate the immune system by attaching to cell receptors called RAGE – Receptors for Advanced Glycation End Products. RAGE ligands are found on endothelial cells, smooth muscle cells, monocytes and monocyte derived macrophages, where they trigger the activation of the inflammatory cascade.

When T1DM patients receive the proper amount of insulin, and blood sugars approach normal, under 120 mg/dl, an interesting thing happens. All of the complications of diabetes are kept in check as well. Micro- and macrovascular complications, cardiac complications, nephrological complications, even periodontal problems tend to return much closer to baseline. Neuropathies, once established, tend to be irreversible, but glucose control has long been recognized as the only way to prevent them or, once they have manifested, to keep them from progressing. This doesn’t mean problems don’t occur – but they no longer occur with the same high frequency as they do in uncontrolled diabetics. As we will see, this is because as glucose and AGE levels trend towards baseline, so does systemic inflammation.

**Inflammation in Type 2 Diabetes**

Type 2 Diabetes mellitus (T2DM) is a more complex, insidious disease than T1DM. As in Type 1 diabetes, there is β-cell death, but the primary reason for this is not autoimmune destruction; it is driven by fatigue and apoptosis. This process does not occur cataclysmically; instead it is usually an insidious process occurring over a period of many months or years as the pancreas gradually loses the ability to produce insulin. Typically, diagnosis is not made until after 40-50% of β-cell function has been lost. Over the natural course of the disease, β-cell function continues to decrease. Analysis of the United Kingdom Prospective Diabetes Study (UKPDS) showed a 4% annual loss, while Bagust and Beale’s analysis of the Belfast Diet Study (BDS) showed that during the natural course of the disease, there is a point where β-cell function begins to drop off at a much steeper rate, as much as 18% annually in uncontrolled diabetics.

In type 2 diabetes, the primary driving factor is insulin resistance. Insulin resistance occurs when cells no longer respond normally to insulin and thus cannot take up glucose in response to cellular needs. The net result is hyperglycemia, as cellular mediators stimulate the liver to release more glucose into the blood stream. In Type 1 diabetes, there are no β-cells, no insulin and so no glucose uptake. In Type 2 diabetes, there is insulin, and cellular uptake of glucose. The process is so compromised that there must be a strong gradient across the cell membrane in order for cells to obtain sufficient quantities.

The most common cause of insulin resistance is obesity. According to the CDC, the combined results of two very large studies covering the period between 1988 and 2002 showed that 85% of T2DM patients were overweight and 54% were clinically obese. Adipose tissue, especially visceral fat, is very highly correlated to insulin resistance and can directly cause insulin resistance through the release of free fatty acids. Free fatty acids (FFAs) stored and released by adipocytes have been shown to increase insulin resistance through the down-regulation of cellular glucose transport activity. This blockade is reversible, so if a patient loses weight, cellular uptake of glucose can improve.

Adipocytes and macrophages associated with visceral fat also synthesize a number of chemicals call cytokines. Many of these cytokines, such as TNF-α, directly and indirectly affect glucose metabolism as well.

**Inflammation in Periodontitis**

Periodontitis begins with the penetration of gram negative, anaerobic bacteria into the gingival sulcus. The bacteria most commonly associated with periodontal disease are Porphyromonas gingivalis, Tannerella forsythia, Aggregatibacter actinomycetemcomitans, Treponema denticola and Prevotella intermedia. Often, more than one of these bacteria are involved in the disease process. For instance, P. gingivalis, T. forsythia and T. denticola are referred to as “red complex bacteria” because they often form complex bacterial mates that stain red when they are fixed to slides.

Of these, P. gingivalis appears to be associated with the greatest systemic response. It has been isolated from tissues in the lungs of patients with pneumonia, in atherosclerotic plaques in blood vessels, circulating in the vascular system and even in a brain abscess. Many of the studies investigating systemic inflammation of periodontal origin measure
the presence of P. gingivalis because of its virulence and its association with so many systemic diseases.

Bacteria that cross the epithelial wall of the gingival sulcus release many toxins that disrupt the epithelial basement membrane and the underlying connective tissue. These toxins include hyaluronidase, collagenase, neuraminidase, catylase, fibrolysin and many more. Toxins make it possible for the bacteria to enter the gingival tissue and disrupt its integrity.

The cell walls of many anaerobes such as P. gingivalis are made of lipopolysaccarides, or LPS. LPS, often called endotoxin in older literature, is very important because it can directly cause inflammation by triggering monocytes and macrophages. In fact, LPS from bacteria often makes its way into the blood stream, where it can trigger a much broader, systemic inflammatory response including the release of many cytokines.

For many years now, we have understood that most of the pathology associated with periodontal disease is caused more by the body’s response to infection than the bacterial infection itself. This is a direct result of the local inflammatory response. When bacteria penetrate the mucosal lining, the first white blood cells on the scene are polymorphonucleocytes, PMNs. These provide the initial immune response by phagocytizing bacteria. In the process, many PMNs undergo apoptosis, and release the contents of their lysosomes, which contain reactive oxygen species (ROS). In lysosomes, superoxides destroy bacteria; in the intercellular matrix, they damage the surrounding tissue, destroying tissue integrity.

PMNs also release Interlukin-6 (IL-6), a proinflammatory cytokine, and Matrix Metalloproteinase-8 (MMP-8) which is a collagenase enzyme. Interluken-6 has many local and systemic effects. One of the most important systemic effects is to dock with Toll-like Receptors (TLRs) on macrophages. This activates the macrophages which begin to churn out a larger set of chemical and messenger compounds, such as cytokines, MMP-1 (another collagenase), and hyaluranidase.

Now there are several collagenases on the scene – collagenases from bacteria, MMP-1 from macrophages and MMP-8 from PMNs. Together, these are the main source of connective tissue destruction. This is why subantimicrobial doses of doxycycline work so effectively to reduce periodontal destruction. At low levels – 20 mg, bid – doxycycline is a potent collagenase inhibitor.

Together, reactive oxygen species, collagenases, hyaluridase and bacterial toxins cause the soft tissue inflammation and destruction typical of the periodontal lesion. But these locally acting factors are not the only proteins released during the inflammatory process. Other cellular products including cytokines have a broader, systemic effect as well.

**Understanding Systemic Inflammation**

There are a wide range of inflammatory diseases, including diabetes, rheumatoid arthritis, Irritable Bowel Syndrome (IBS) and periodontal disease. Obesity, can also be considered an inflammatory disease. Inflammatory diseases create and respond to a class of protein messengers called proinflammatory cytokines. In diabetes, as we have discussed, inflammation arises from hyperglycemia, AGEs and free fatty acids. In rheumatoid arthritis, inflammation is in response to joint destruction. In periodontal disease, inflammation develops in response to bacterial invasion. By whatever pathway inflammation arises, the inflammatory compounds and messengers are very much alike. Nature is, after all, very thrifty. The next sections discuss three of the most important proinflammatory cytokines responsible for bone loss and examines how they affect periodontal disease and diabetes. Table 1 provides a summary of this information.

<table>
<thead>
<tr>
<th><strong>Periodontitis</strong></th>
<th><strong>Diabetes</strong></th>
<th><strong>Autocrine</strong></th>
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<tbody>
<tr>
<td><strong>TNF-α</strong></td>
<td>PMN chemo-attractant</td>
<td>Blocks insulin receptors leading to insulin resistance</td>
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<tr>
<td>Stimulates macrophages to produce cytokines</td>
<td>Mobilizes adipocyte lipids</td>
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<td>Stimulates osteoelastic activity</td>
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<tr>
<td><strong>IL-1β</strong></td>
<td>Capillary wall permeability</td>
<td>Capillary wall permeability</td>
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<td>Stimulates collagenase production</td>
<td>Directly causes β-cell death</td>
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<tr>
<td>Stimulates osteoelastic activity</td>
<td>Stimulates liver to produce CRP and complement</td>
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<tr>
<td><strong>PGE2</strong></td>
<td>Small vessel dilation</td>
<td>Small vessel dilation</td>
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<td>Stimulates osteoclast differentiation</td>
<td>Contributes to adipose tissue formation</td>
<td>Regulates adipocyte lypolysis and leptin release</td>
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<td>Marker for adipose tissue inflammation</td>
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Table 1
**Tumor Necrosis Factor-α (TNF-α)**

In periodontal disease, monocytes and monocyte-derived macrophages are recruited to inflamed tissue by PMNs and local inflammatory factors where they release several cytokines, including TNF-α. TNF-α exerts several local effects. First, it is one of several cytokines that recruit white blood cells to an area of inflammation. TNF-α is a powerful chemo-attractant for PMNs.18

TNF-α signals macrophages to produce IL-6, which in turn stimulates the liver to produce many acute phase inflammatory peptides, including complement, fibrinogen and high-sensitivity C-reactive protein (CRP).19 These compounds play an important role in many diseases, including arteriosclerotic heart disease and stroke. TNF-α has been associated with other inflammatory diseases, particularly rheumatoid arthritis, where anti-TNF-α therapy is being tested to control the disease.

TNF-α has also been shown to be a powerful stimulator of osteoclastic activity.16 It is the first of several peptides we will examine that are responsible for the bone loss associated with periodontal disease. Studies have shown an increase in TNF-α in gingival crevicular fluid (GCF) and systemically in blood serum associated with periodontal disease. Engebretson showed a log-linear relationship between attachment loss and serum TNF-α levels.20 This is an important finding, because it not only shows that TNF-α levels are associated with periodontal bone loss, but that the magnitude of the bone loss correlates with the serum levels found. Another study by Andrukhov and his colleagues demonstrated that subjects with generalized periodontitis showed an increase in serum TNF-α levels associated with periodontal disease. Studies have shown that both TNF-α and IL-1β are capable of causing osteoclasts to differentiate and leads directly to net alveolar bone loss.23

In contrast to other cytokines like TNF-α and CRP, Chen et al found that serum concentrations of IL-1β do not appear to rise as periodontal disease becomes more severe. However, tissue and GCF levels of IL-1β and IL-6 do change, correlating strongly with periodontal disease severity.24

In diabetes, IL-1β is released directly from adipocytes as well as macrophages found in visceral fat deposits. Under inflammatory conditions, IL-1β in adipose tissues upregulate the cyclooxygenase-2 (COX-2) pathway that leads to the creation of PGE_2 (see below).

IL-1β is especially significant because it can directly cause β-cell death in the pancreas.25 Indirectly, IL-1β increases insulin resistance by stimulating the liver to release CRP and other acute phase products such as fibrinogen and complement, although this effect is not as strong as that seen with IL-6. In addition to the systemic effects noted above, many of the acute phase products such as CRP, can influence insulin resistance.

**Interlukin 1β (IL-1β)**

Interlukin 1β is the classic pyrogen. It can cause fever systemically and the warmth associated with infection locally. In periodontal disease, it is produced by both PMNs and macrophages.

In the periodontal lesion, IL-1β has many important effects. The redness in periodontal disease is due in part to the capillary wall permeability that occurs in response to this cytokine. IL-1β stimulates collagenase production, which in turn breaks down periodontal connective tissues. Most importantly, it leads to the formation of osteoclasts and concomitant bone loss. Studies have shown that both TNF-α and IL-1β are capable of causing osteoclasts to express a protein called Receptor Activator for Nuclear factor-kappa Beta Ligand or RANKL. RANKL stimulates precursor osteoclasts to differentiate and leads directly to net alveolar bone loss.23

**Prostaglandin E_2 (PGE_2)**

PGE_2 is another cytokine released by activated monocytes, macrophages and PMNs in gingival tissues. In the periodontal lesion, PGE_2 increases small vessel dilation, allowing for a greater exchange of leukocytes, lymphocytes and cytokines. PGE_2 has been shown to strongly influence osteoclastic activity. During physiologic bone repair, PGE_2 is secreted by osteoblasts. In this it acts as an “autocrine” – a peptide used as part of a control or feedback loop in the system that produces it.26 This is similar to the way TNF-α functions in healthy adipose tissues. As you can see, nature is thrifty.

In periodontal disease, exogenous PGE_2 is considered one of the key cytokines that cause alveolar bone destruction.
In fact, according to Noguchi and Ichikawa,\(^2\) the RANKL pathway through which IL-1\(\beta\) acts to destroy alveolar bone is mediated by PGE\(_2\), production.

In diabetes, adipocytes are the major source of PGE\(_2\), where it is part of the regulatory pathway down-regulating lipolysis and increasing leptin release.\(^{28}\) Decreased lipolysis results in increased adipose tissue mass,\(^{29}\) while leptin signals the hypothalamus to decrease appetite. In obesity, the PGE\(_2\), regulatory role becomes overwhelmed by other hormonal and inflammatory effects. Leptin and serum PGE\(_2\), levels increase, adipose tissue mass increases and the hypothalamus becomes leptin resistant, so appetite is no longer suppressed. Serum PGE\(_2\), levels are regarded as a good measure of the level of systemic inflammation resulting from obesity.

So now that we know a little about a few of the major cytokines involved in inflammation, what do we know about the ability of cytokines like TNF-\(\alpha\) that originate from periodontal tissues to affect insulin receptors? Can IL-\(\beta\) generated in visceral fat deposits influence the course of periodontal bone loss? The next sections will show how mediators created in one disease can influence the other.

**Diabetic Effects on the Periodontium**

The effects of diabetes on periodontal disease has been one of the most studied systemic interrelationships in the last 20 years. Epidemiologic studies like one by Tsai using data from the National Health and Nutrition Examination Study III (NHANES III), a large medical study conducted from 1988 to 1994, showed an almost three-fold increase in periodontal disease among uncontrolled diabetics.\(^{3}\) Other studies have suggested rates of periodontal disease in uncontrolled diabetic cohorts as high as 6 times that found in the non-diabetic population.\(^{30}\)

It has only been relatively recently that studies have begun to describe the influence of the classic diabetic intermediaries on periodontitis. Just as AGE/RAGE interactions take center stage in vascular and neurological diabetic complications, it appears that these compounds are also at the heart of diabetes’ influence on the course of periodontal disease. Studies have shown that AGE can affect gingival epithelial cell integrity and can alter collagen synthesis, reorganization and turnover.\(^{31}\) Collagen modified by AGEs can accumulate in blood vessel walls, thickening the walls and decreasing vessel lumens. In large blood vessels, AGEs can also cause up-regulation in cellular adhesion molecules and alter LDL, making them more likely to adhere to the endothelium. This contributes to increased atheroma formation.\(^{4}\) Under the influence of AGEs, PMNs become less motile and more subject to apoptosis and thus release more ROS into the connective tissue milieu.

Monocytes and macrophages have RAGE ligands and become hyper-reactive in the presence of AGE. This causes them to release far more inflammatory peptides such as TNF-\(\alpha\) than granulocytes not subject to AGE interactions. Salvi, Beck and Offenbacher found that in response to a challenge from P. gingivalis-derived LPS, monocytes from diabetic patients secreted four times the amount of PGE\(_2\), IL-1\(\beta\) and TNF-\(\alpha\).\(^{32}\)

RAGE mediated granulocyte hyper-reactivity is probably the single largest effect that either disease has on the other and its importance cannot be overstated. Lalla and his colleagues illustrated the importance of RAGE’s influence on alveolar bone destruction. They showed that blocking AGE receptors effectively reduced alveolar bone loss associated with P. gingivalis in a controlled experiment on mice.\(^{33}\)

The relationship is more complicated in type 2 diabetics. Adipose tissues release cytokines such as TNF-\(\alpha\) and IL-6 that affect periodontal tissues. The systemic level of these cytokines can be as greatly influenced by obesity as they can by hyperglycemia and so overweight individuals with adequately maintained glucose levels may still have high levels of circulating cytokines that can affect periodontal disease progression. This effect may serve to reduce the statistical relationship between glucose levels and periodontal disease, underestimating the true impact of hyperglycemia and RAGE/AGE on periodontal tissue destruction. For instance Khanna and Mali showed a set of interrelated correlations between obesity, periodontal disease and TNF-\(\alpha\). They found that BMI and probing depths were strongly related. Both TNF-\(\alpha\) and probing depths and BMI and TNF-\(\alpha\) were also related,\(^{34}\) suggesting that increased TNF-\(\alpha\) associated with obesity may be influencing periodontal disease and pocket depth. Ramirez-Torosa, et al also found close associations between periodontitis, plasma fatty acid profiles and metabolic risk factors such as TNF-\(\alpha\) and soluble vascular cell adhesion protein (VCAM),\(^{35}\) which has been associated with atherosclerosis.

**Periodontal Disease and Glucose Control**

Relatively few studies have investigated the increased risk of diabetes development among patients with periodontal disease. In an analysis of NHANES III data, Soskolne and Klinger show that 12.5% of subjects with periodontal disease had diabetes, compared to only 6.3% of subjects without periodontal disease.\(^{36}\) In contrast, Ide, et al concluded that after isolating the impact of periodontal disease on diabetes by accounting for major confounders, periodontal disease did not increase diabetes incidence.\(^{37}\) Using a prospective database of Japanese workers, they showed that while unadjusted analysis showed a significant correlation between the two diseases, after adjusting for BMI, hyperlipidemia, hypertension and other major co-factors, periodontal disease did not appear to predispose subjects to increased diabetes risk.

On the other hand, many studies have investigated the effect that periodontal disease has on diabetes. Taylor\(^{38}\)(2001) and Taylor and Borgnakke\(^{39}\) (2008) reviewed studies on the effects of periodontal disease and glucose control, finding that...
the majority of studies demonstrated a correlation between periodontitis and hyperglycemia.

While most studies divided subjects into discrete groups and calculated relative risks, a few studies plotted glucose levels vs. continuous variables. In 2001, Grossi showed that in a cohort of Pueblo Indians, HbA1c levels were correlated with the percentage of probing sites higher than 5mm. See Figure 1. Nesse and his colleagues also found a similar relationship in percentage of probing sites higher than 5mm.40 See Figure 1. In a cohort of Pueblo Indians, HbA1c levels were correlated with the percentage of probing sites higher than 5mm.40 See Figure 1. Nesse and his colleagues also found a similar relationship in percentage of probing sites higher than 5mm.

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Figure 1. Glycated Hemoglobin vs. Probing Depth

![Glycated Hemoglobin vs. Probing Depth](image)


The causal relationship between periodontal disease and hyperglycemia is not as well established. While several studies have shown significant reduction in HbA1c levels following scaling and root planing (S/RP), with or without antibiotics, other studies have failed to confirm these findings. Part of the problem lies with the studies themselves. There is no standard definition of “moderate” or “severe” gum disease. Often patients with a few isolated 5 or 6 millimeter pockets get lumped together with patients who have many infected sites or significant, generalized bone loss. This can wash out study results and lead to marginal statistical significance.

Many clinical studies have small sample sizes which limits statistical inference. This is especially true for systemic studies. There are many environmental and physical factors that can influence the absolute levels of any systemic marker we may choose to measure. Since the changes studies are trying to elucidate are quite modest, many subjects are required to achieve the statistical power to overcome the background noise that these other influences create.

As the quality and sophistication of perio-systemic studies continues to improve, it is becoming evident that as periodontal inflammation becomes more generalized, systemic consequences become more significant. Certainly it makes sense that an isolated 5mm pocket with moderate bleeding on probing will have a much smaller effect than generalized type IV periodontitis with advanced bone loss. Indeed, the few studies, like Grossi’s that correlate periodontal disease as a continuous variable against glucose levels show that the number of periodontally involved sites is important.

**Periodontal Disease and Systemic Inflammation**

So now we know that several cytokines have an effect on periodontal tissues, β-cells and insulin receptors and that these cytokines are created and released in both diseases. But what is the relative systemic impact of the two diseases? Certainly we are all aware of diabetes’ devastating effects on vascular and neurological systems. Can periodontal disease contribute to these problems through its influence on circulating cytokines?

Clinically, we divide periodontal diseases into 4 stages, depending on the level of disease. The systemic impact may differ as periodontal disease progresses from gingivitis to severe bone loss and chronic inflammation. Bleeding on probing (BOP), a measure of soft tissue inflammation, can vary significantly over the course of the disease. This is especially true in patients who smoke and have periodontal disease. Bleeding may be significantly lower in a type IV periodontal patient than one with no bone loss, who only has gingivitis. There is some evidence that the systemic response to these varying disease states has different systemic implications. Beck shows that BOP is highly correlated to serum soluble intracellular adhesion molecule (sICAM) levels, while pocket depth is more correlated to CRP levels. sICAM is an important peptide associated with atherosclerotic plaque development. Marcaccini demonstrated that periodontal disease was associated with higher plasma levels of MMP-3, MMP-8 and MMP-9. Treating the disease with scaling and root planing showed statistically significant reductions in MMP-8 and 9.

Many studies have shown higher levels of CRP, an acute phase peptide produced by the liver in the presence of IL-6 or TNF-α. Gomes-Filho, et al found a significant correlation between the presence of periodontal disease and CRP in a cohort of 359 subjects. CRP has been implicated in the creation of atherosclerotic plaques and has been found to correlate well with the risk of cardiac events such as acute cardiac syndrome and stroke. In a 2008 review of literature, Persson & Persson found that the majority of studies they reviewed reported that periodontal disease was associated with increased cardiovascular disease and CRP levels.

Plasma levels of many cytokines have been shown to rise with increasing severity of periodontal disease. Significantly, several studies, like one by Engebretson, show that cytokine levels often correlate better with bone loss or attachment loss than with BOP levels or pocket depth. This relationship appears to hold true in other populations than diabetics. The review by Persson and Persson reported...
that alveolar bone loss correlates better than BOP or pocket depth with cardiovascular disease.

Studies on rheumatoid arthritis (RA) show that TNF-α is a major contributor to joint inflammation. In a study of rheumatoid arthritis patients with periodontal disease, Ortiz showed that periodontal treatment significantly reduced joint tenderness and swelling. Lower serum TNF-α concentrations were associated with reduction of RA inflammation. Interestingly, TNF-α blocking agents used to treat rheumatoid arthritis were responsible for highly significant positive changes in clinical attachment levels and gingival index values.

The bacteria associated with gingivitis and early periodontal disease differs from the more aggressive anaerobic bacteria like P. gingivalis and A. actinomycetemcomitans found in deep pockets. It is the latter bacteria that have been associated with systemic bacteremia, systemic endotoxin (LPS) levels and acute inflammatory peptide production.

Controlling Hyperglycemia May Reduce the Progress of Periodontal Disease

Just as maintaining good glycemic control has been found to control damage to vascular and neurological systems, maintaining glycemic control can reduce the rate of periodontal destruction in periodontitis. As we have seen, hyperglycemia causes an increase in AGE/RAGE and subsequently an increase in many of the proinflammatory cytokines found in inflamed gingival tissues. The equilibrium between osteoblasts and osteoclasts is shifted towards clastic activity, leading to more rapid destruction of alveolar bone. Highly significant correlations between glycemic levels and levels of TNF-α, IL-1β and other cytokines in gingival crevicular fluids have been established.

Studies that have compared bone loss in controlled vs. uncontrolled diabetic subjects have shown there is significantly more destruction in the uncontrolled diabetic group. In 1986, Tervonen and Knuuttila studied 103 subjects that showed poor glycemic control was correlated to increased numbers of periodontal pockets. This result was confirmed by two later studies. In their landmark 1998 prospective study, Taylor, et al showed that subjects with HbA1c levels at 9% or higher had significantly more alveolar bone loss over the study’s duration than better controlled diabetics. In 2002, Tsai et al in their evaluation of cross-sectional NHANES III data, were able to show that this relationship could be observed in a large population.

The bottom line is that controlling diabetes may have a very significant impact on the progression of periodontal disease. Achieving good glycemic control decreases the level of AGE products. Since AGE has been shown to make macrophages hyper-responsive, reducing AGE/RAGE values also reduces the level of proinflammatory cytokines in gingival tissues that stimulate osteoclastic activity. Even in the presence of active periodontal disease, this may reduce the extent and speed of osseous destruction.

Weight loss for the nearly 90% of type 2 diabetics that are overweight can also reduce the levels of TNF-α and IL-1β that promote bone loss. While weight loss reduces AGE levels through its affect on insulin resistance and hyperglycemia, to some extent, these two factors are independent. Each of the three cytokines discussed above that affect bone loss are reduced by losing weight, potentially shifting the equilibrium between bone creation and bone loss away from osseous destruction.

Periodontal Therapy May Result in Better Glycemic Control

Because the systemic effect of periodontal disease is smaller than that of diabetes, the impact of periodontal disease on diabetes is more subtle. As we have seen, the majority of studies suggest that active periodontal disease is correlated with increased risk for diabetes, but the magnitude of this effect has not been definitively established. Nonetheless, when the impact of periodontal treatment on systemic biomarkers or on blood glucose levels are investigated, research indicates there is a significant positive result.

When the inflammation of active periodontal disease is reduced through S/RP, levels of circulating cytokines are also reduced. Borkhar showed that S/RP reduced levels of fibrinogen, CRP and white blood cells in a study of CHD patients. Sun, demonstrated that S/RP resulted in reduced levels of CRP, TNF-α and IL-6 three months after therapy in T1DM and impaired glucose tolerance (IGT) patients. Lalla, et al found that full-mouth debridement reduced serum levels of CRP and macrophage secretion of TNF-α. Several of the CHD studies reviewed by Persson & Persson also showed significant reduction in serum biomarkers such as CRP, IL-6 and TNF-α after periodontal therapy.

Even in subjects without diabetes, circulating cytokine levels may be reduced through scaling and root planing. Shimada et al report that periodontal treatment reduced levels of IL-6 and CRP in a cohort of periodontally involved non-diabetic subjects.

Several studies confirm that S/RP treatment can have a positive effect on diabetes. In a meta-analysis of current studies, Darré et al concluded that the weight of evidence showed that S/RP could produce significant reduction in A1c levels. Their meta-analysis predicted an average A1c reduction of 0.6%. This is significant, because the reduction in A1c from many of the second or third tier diabetes medications averages 1.0 to 1.5%.

Adjuncts to scaling and root planing do not appear to further reduce A1c levels, even though they often lead to better resolution of the periodontal disease. For instance, O’Connell, Tabu and colleagues showed that while S/RP plus doxycycline 20 mg bid lowered A1c levels more than S/RP alone, the difference was not significant. Doherty’s group showed the same result with minocycline microspheres (Arestin).

The sample size of both studies was...
small, so larger studies may show modest but significant differences do indeed exist.

The ability of S/RP to affect A1c levels may be independent of glycemic control. Three studies\(^5\) divided diabetic subjects into groups based on A1c levels. While all of the studies showed A1c reductions in both subgroups, only the most recent, by Ou and Li, was large enough to show significant changes in both well controlled and poorly controlled diabetic subgroups. This study is important because it showed that the magnitude of the reduction in A1c was nearly identical for both subgroups (0.45 vs 0.47\%).

One of the most important results of these studies is that the level of metabolic control did influence the effectiveness of periodontal therapy. While S/RP statistically improved periodontal measures such as BOP and probing depths for all participants (controls and diabetics), the better controlled diabetic subgroup often had a better overall periodontal responses to treatment. Kardeşler\(^7\) showed that after periodontal therapy, BOP scores were lower in all groups, but BOP remained at a statistically higher level in the poorly-controlled diabetic subgroup.

Predicting the Effects of Periodontal or Diabetes Intervention

Periodontal disease and diabetes mellitus are both inflammatory diseases. They induce a variety of tissues including endothelial cells, fibroblasts, adipocytes, monocytes and macrophages to release a cascade of peptides called cytokines. Cytokines exhibit a variety of local and systemic actions. Some actions, like TNF-\(\alpha\) expression in adipocytes, are involved in physiologic regulatory functions. In other cases, such as when TNF-\(\alpha\) is released from macrophages associated with visceral fat, the actions are proinflammatory. Typically, the function of the cytokine depends on the target tissue. In the pancreas IL-1\(\beta\) can cause apoptosis of \(\beta\)-cells; in periodontal tissues, it causes osteoclast differentiation and bone loss.

Increasing severity of either disease is associated with increases in cytokine expression. Levels of important cytokines like IL-1\(\beta\), TNF-\(\alpha\) and PGE\(_2\) in serum, GCF and periodontal or adipose tissues have all been shown to correlate with increasing diseased states. This may explain the strong two-way relationship between the diseases. Severe periodontal disease can lead to poor glycemic control; poorly controlled diabetes can increase the severity of alveolar bone loss.

More recently, research has also shown that the relationship holds in the other direction as well. As diabetes is brought under control, cytokine and AGE levels are also reduced. A reduction in the severity of periodontal inflammation and bone loss can also occur. Scaling and root planing affects glycemic control. Recent analysis has suggested that S/RP can reduce average HbA1c readings by approximately 0.6%.

Can these results be used clinically to predict changes in glycemic control or periodontal health in our practices? Presently, the answer is “No.” All of the studies reviewed above show too much variation to allow clinicians to be able to tell their patients that treating their periodontal disease will result in lower A1c levels or reduce the number of the medications they are taking. Likewise, bringing hyperglycemia under control in a newly-discovered diabetic patient cannot guarantee that their bleeding scores will go down or that their rate of bone loss will be reduced.

But the information above should allow clinicians to confidently state that intervention strategies are likely to help their patients and to reinforce recommendations for necessary treatment. Patients can be told that the more generalized or severe their periodontal disease is, the more likely it is to adversely affect their diabetes. Grossi and Nesse both show that one or two 5mm pockets in an otherwise healthy mouth will have less of an effect on diabetes than generalized 6-7 mm pocketing with substantial attachment loss. At the same time, patients should be advised that while studies like the one by Ou and Li suggest S/RP is as likely to modestly reduce hyperglycemia in the poorly-controlled diabetic as in the well controlled individual, therapeutic results may be compromised.

For the same reasons, we cannot tell our diabetic patients that controlling hyperglycemia will eliminate their periodontal disease. Patients can be advised that in order to maximize the value of their periodontal treatment, they need to keep their blood sugar levels under tight control. Well-controlled diabetics may heal better – Kardeşler showed that BOP, pocket depths and clinical attachment loss after S/RP were all lower in the controls and better-controlled diabetics. Patients need to understand that periodontal disease is a chronic, progressive disease; periodontal disease is not cured, it is arrested. Well-controlled diabetics may experience a slower pace of alveolar bone loss than their poorly-controlled peers.

Lowering blood sugars may not be enough to influence periodontal disease progression or the results of periodontal therapy in the 85% of type 2 diabetics that are overweight. While controlling blood sugars can reduce the effect of AGE on collagen destruction and macrophage hyper-reactivity, obesity alone can result in levels of TNF-\(\alpha\) and IL-1\(\beta\) high enough to contribute to alveolar bone destruction.

At the most basic level, we can tell our patients that nature is thrifty. The same mechanisms that result in bone loss can cause \(\beta\)-cell destruction and insulin resistance. Cytokines like TNF-\(\alpha\), IL-1\(\beta\) and PGE\(_2\) are the common destructive thread in inflammatory diseases like periodontitis and diabetes.

References

3. Goldin A, Beckman JA, Schmicit AM, Greager MA.
Advanced Glycation End Products: Sparking the Development of Diabetic Vascular Injury


24. Salvi GE, Beck JD, Offenbacher S. PGE2, IL-1 beta, and TNF-alpha responses in diabetics as modifiers of periodontal disease expression.


26. Ramirez-Torosa MG, Quiles JL, Battino M, Granados S, Morillo JM, Dompardre S, Newman HN, Bullon P. Periodontitis is associated with altered plasma fatty acids...


Author Profile
Jeffrey A. Sibner, D.M.D. is an Adjunct Assistant Professor at the University of Pennsylvania School of Dental Medicine where he teaches a course on periodontal disease and diabetes. He is affiliated with St. Mary Medical Center and teaches a lay course called Diabetes and You through their diabetes education program. Dr. Sibner maintains a private practice in general dentistry in Bucks County, Pennsylvania.

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1. Which of the following is NOT an inflammatory disease:
   a. Diabetes
   b. Rheumatoid Arthritis
   c. Irritable Bowel Syndrome
   d. Pneumonia

2. Which of the following statements is TRUE:
   a. AGEs are produced primarily in the liver
   b. RAGE ligands on macrophages mediate a hyper-reactive immune response
   c. Glycation of proteins and lipids to form AGE is enzymatically mediated
   d. AGEs are only produced in people with diabetes

3. Which of the following diseases are NOT affected by periodontal disease?
   a. Atherosclerosis
   b. Stroke
   c. Obesity
   d. None of the above

4. In poorly controlled diabetic patients, periodontal therapy
   a. Doesn’t work
   b. May be less effective
   c. Is just as effective
   d. Should never be attempted

5. Lipopolysaccharides from anaerobic bacteria like P. gingivalis do all of the following:
   a. Initiate gingival inflammation
   b. Cause most periodontal bone loss
   c. Cause a variety of systemic effects
   d. All of the above

6. TNF-α is a cytokine that exhibits which of the following actions:
   a. Increases osteoclast differentiation resulting in alveolar bone loss
   b. Is a chemo-attractant for PMNs
   c. Acts as an “autocrine” in adipocyte metabolism
   d. All of the above

7. In Type 1 diabetes, systemic damage is caused primarily by:
   a. AGE/RAGE reactions
   b. Ketoacidosis
   c. Free fatty acids (FFAs)
   d. PGE

8. In one study, S/RP did not affect serum concentrations of which cytokine?
   a. TNF-α
   b. PGE
   c. IL-1β
   d. CRP

9. Which measure correlated best with circulating cytokines:
   a. Periodontal therapy on patients with poorly controlled diabetes
   b. Reducing A1c levels to below 7 or 8% will reduce alveolar bone loss
   c. Gingival hypertrophy
   d. PGE, in gingival crevicular fluid

10. Based on our current understanding of the relationship between periodontitis and diabetes, clinicians can accurately make which of the following statements to their patients:
    a. Periodontal therapy on patients with poorly-controlled diabetes may not result in adequate healing
    b. Reducing A1c levels to below 7 or 8% will reduce pocket depths
    c. Periodontal therapy will lower A1c by 0.6%
    d. Patients with type 1 or Type II periodontal disease that have scaling and root planing will lower their A1c levels

11. S/RP may reduce A1c levels in which patients:
    a. Poorly-controlled diabetics (A1c > 8%)
    b. Well-controlled diabetics (A1c ≤ 8%)
    c. Both groups
    d. Neither group

12. Which cytokine is NOT released by macrophages or PMNs?
    a. CRP
    b. IL-1β
    c. TNF-α
    d. All of the above

13. In visceral fat, serum TNF-α is derived primarily from:
    a. Adipocytes
    b. Macrophages
    c. Lipocytes
    d. All of the above

14. Which cytokine does NOT function as an autocrine?
    a. TNF-α
    b. PGE
    c. IL-1β
    d. Both A and C

15. Collagenase is an important enzyme in periodontal disease because it breaks down soft tissue. Which of the following secretoregulyse collagenases?
    a. T. forsythia
    b. PMNs
    c. Macrophages
    d. All of the above

16. In Type 2 diabetes, diagnosis is typically made after what percentage of β-cell function is lost?
    a. 80%
    b. 45%
    c. 50%
    d. 70%

    a. They are both inflammatory diseases
    b. Small sample sizes may affect the ability to show statistical significance
    c. Europeans may have more resistant periodontal disease
    d. Type 2 diabetes is caused by insulin resistance; β-cells are not affected

18. What factors may influence the effectiveness of periodontal therapy in Type 2 diabetes?
    a. Fasting blood glucose levels
    b. Obesity
    c. Oral hygiene
    d. All of the above

19. Prostaglandin E2 (PGE2) has all of the following functions:
    a. Acts as an “autocrine” in adipocyte metabolism
    b. Is a chemo-attractant for PMNs
    c. Increases osteoclast differentiation resulting in alveolar bone loss
    d. All of the above

20. Diabetes and periodontal disease exhibit many common systemic traits because:
    a. They are both inflammatory diseases
    b. They release the same kinds of proinflammatory cytokines
    c. PGE, and TNF-α exhibit identical actions in both diseases
    d. All of the above

21. AGE is an acronym for:
    a. Age-Generated Erythrocytes
    b. Adjusted Glycemic Evaluation
    c. Advanced Glycogen Enzymes
    d. Advance Glycation End products

22. Receptors for AGE (RAGE) can be found on all of the following EXCEPT:
    a. Endothelial cells
    b. Collagen
    c. Smooth muscle cells
    d. Monocytes

23. In T1DM, the rate of most diabetic complications is reduced to near-normal when glucose levels are well controlled. Which is the exception to this rule?
    a. Kidney disease
    b. Neurologic complications
    c. Retinopathy
    d. Vascular diseases

24. The periodontal pathogen most associated with systemic disease is:
    a. P. gingivalis
    b. T. forsythia
    c. P. intermedia
    d. T. Denticola

25. The white blood cells that form the initial line of defense against bacterial invasion into periodontal tissue are:
    a. Monocytes
    b. T cells
    c. PMNs
    d. Monocyte-derived macrophages

26. Proinflammatory cytokines can directly and indirectly stimulate the liver to release which of the following acute phase inflammatory peptides?
    a. Fibrinogen
    b. Compliment
    c. CRP
    d. All of the above

27. PGE, is released by all of the following EXCEPT:
    a. Endothelial cells
    b. Monocytes
    c. Adipocytes
    d. Osteoblasts

28. PGE, can exhibit the following functions:
    a. Bone repair
    b. Bone destruction
    c. Leptin reuptake
    d. Leptin release

29. Periodontal disease affects diabetes, and diabetes influences periodontal disease. According to Lalla, which has the largest effect?
    a. CRF-mediated increase in coronary vascular disease
    b. RAGE-mediated hyper-responsiveness in macrophages
    c. Adipocyte-mediated increase in PGE, bone destruction
    d. Lipopolysaccharide-mediated inflammation of endothelium

30. Periodontal disease and diabetes have profound commonalities. In our clinical practices we should:
    a. Tell our diabetic patients that studies show they will lower their blood sugars by treating active periodontal disease.
    b. Tell our diabetic patients that because studies do not always show that treating periodontal disease can reduce A1c levels, there is no established clinical relationship between the two diseases.
    c. Tell our diabetic patients with generalized periodontal disease that studies show periodontal treatment has led to statistically significant, modest reductions in A1c levels, and they may find their A1c levels go down after periodontal therapy.
    d. Tell our diabetic patients with isolated 6mm pockets that studies show periodontal treatment has led to statistically significant, modest reductions in A1c levels, and they may find their A1c levels go down after periodontal therapy.
The Inflammatory Origins of Periodontal Disease and Diabetes: A Framework for Understanding Clinical Outcomes

Educational Objectives
1. The role of inflammatory cytokines in periodontal bone destruction and insulin resistance in diabetes.
2. The effect of periodontal disease on blood sugar levels.
3. How to advise patients and predict treatment outcomes for peri-patients with diabetes.

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2. To what extent were the course objectives accomplished overall?  5 4 3 2 1 0
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