Periodontitis, Diabetes Mellitus, Cardiovascular Disease
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Over the last decades, the relation between periodontitis, diabetes mellitus (DM) and atherosclerotic cardiovascular disease (ACVD) has been extensively studied. Periodontitis is a common chronic multifactorial inflammatory disease of the supporting structures of the teeth and is a major cause of tooth loss (Pihlstrom et al., 2005). Besides the possibility that systemic diseases, such as DM (Preshaw et al., 2012), may contribute to the onset and/or progression of periodontitis, inflamed periodontal tissues open the possibility for oral bacteria to enter the circulation and induce systemic inflammation. It is now widely accepted that a low grade systemic inflammation contributes to metabolic dysregulation (King, 2008, Pradhan et al., 2001) and atherogenesis (Kaptoge et al., 2010, Friedewald et al., 2009). Although many investigations suggest several pathways that might explain the link between periodontitis and DM, and periodontitis and ACVD, the biological mechanisms, clinical relevance and implications of these interactions are still not well understood. The aim of this thesis was to contribute to the understanding of the complex relationship between these three disease states intricately caught in a ‘Bermuda Triangle’.

PERIODONTITIS AND DIABETES MELLITUS

DM and periodontitis are two chronic diseases that have long been considered to be biologically linked in a bi-directional way (Llambes et al., 2015, Preshaw et al., 2012, Lalla and Papapanou, 2011) (Figure 1).

*Figure 1* The bi-directional link between diabetes mellitus and periodontitis. First, periodontitis is considered as a complication of diabetes mellitus. Second, periodontitis, as a chronic inflammatory disease, might contribute to an increased insulin resistance.

**The DM → periodontitis connection**

Already in the nineties, periodontitis has been considered as complication of DM by the former director of NIH/NIDCR (Løe, 1993) (Figure 1). Thus, the initiation and progression
of periodontitis could be indicative for metabolic dysregulation, and it has been suggested that the dental setting might be suitable for diabetes screening (Lalla and Lamster, 2012).

Since recently plasma glycated hemoglobin (HbA1c) levels were proposed as a suitable new marker for DM screening (American Diabetes Association, 2015) and a new validated chairside-method for HbA1c testing was available, the hypothesis could be tested, that periodontitis patients show an increased prevalence of suspected (pre)diabetes compared to subjects without destructive periodontal disease (Chapter 2). Therefore, 126 mild/moderate periodontitis patients, 78 severe periodontitis patients and 109 subjects without periodontitis were screened for DM, using the finger stick method and a validated HbA1c dry spot analysis. Applying the ADA-guidelines for DM diagnosis (American Diabetes Association, 2015), there was a significant overrepresentation of subjects with suspected diabetes and prediabetes in the severe periodontitis group and the mild/moderate periodontitis group, compared to the control group. Notably, 18.1% suspected new diabetes patients (in addition to 7.7% already diagnosed) were found among subjects with severe periodontitis compared to 9.9% and 8.5% in subjects with mild/moderate periodontitis and controls, respectively. This confirms the assumption that severe periodontitis could be one of the first signs of metabolic dysregulation and undiagnosed DM and that the dental office, with particular focus on periodontitis patients, proved to be a suitable location for screening for (pre)diabetes (Figure 2).

Early diagnosis and intervention of (pre)DM prevent the common micro- and macro-vascular complications (Tian et al., 2014) and are cost-effective, by delaying or preventing the direct medical costs of diabetes, including the costs of diabetes education and nutritional counseling, glucose monitoring, treatment, surveillance for complications, and treatment of complications (Herman et al., 2005). Owing to the absence of symptoms and/or disease-related knowledge, DM often goes undetected, and approximately half of people with DM are not aware of their status (Lee and Colagiuri, 2016). Therefore, risk indicators for (pre)DM screening are needed and proposed (American Diabetes Association, 2015). It is proposed here to include screening for DM in the dental office, with particular focus on periodontitis. When a suspected case of DM is found, the definitive diagnosis is made by the family physician and treatment can be initiated. Reciprocally, this is also relevant for the successful treatment of periodontitis (Eke et al., 2016, Garcia et al., 2015) (Figure 1, 2). It can be expected that, in the future, a good collaboration between dentists and diabetologists will lead to the best periodontal and diabetes care.

The periodontitis → DM connection
There is a growing body of evidence supporting the fact that periodontal inflammation adversely affects glycemic control (Engebretson and Kocher, 2013, Preshaw et al., 2012).

226
One biological explanation might be the increased pro-inflammatory state due to periodontitis (Schenkein and Loos, 2013), resulting in increased insulin resistance (King, 2008, Shoelson et al., 2006) (Figure 1). A systematic review (Chapter 3) presented in this thesis was designed to explore the robustness of observations that periodontal treatment affects glycemic control in patients with type 2 DM. Screening of the initially 639 identified studies, using two online databases, resulted in 5 suitable papers. A total of 371 patients were included in this analysis, with periodontitis as predictor and the actual absolute change in HbA1c ($\Delta$HbA1c) as the outcome. The duration of follow-up was 3 to 9 months. All studies described a research population with both periodontitis and type 2 DM, in which glycemic control improved after periodontal therapy compared with a periodontitis non-intervention control group (i.e. no periodontal treatment). A meta-analysis, evaluating $\Delta$HbA1c before and after periodontal therapy, demonstrated a reduction of 0.40% in treatment groups favoring periodontal intervention, suggesting that periodontal treatment leads to an improvement of glycemic control in patients with type 2 DM for at least 3 months (Figure 2). The observed decrease of HbA1c is also clinically relevant, since any decrease of HbA1c will result in less diabetic complications (Genuth et al., 2003, Unger, 2008).

Although results of several more recent systematic reviews and meta-analyses corroborate the findings of Chapter 3, the magnitude of reduction in HbA1c after periodontal treatment varies (Engebretson and Kocher, 2013, Mauri-Obradors et al., 2015, Sgolastra et al., 2013, Simpson et al., 2015, Teshome and Yitayeh, 2016), indicating that the external validity might not be so high. This is also demonstrated by a recent, large multi-centered single-blind, randomized controlled trial, which showed no differences in HbA1c values between periodontally treated and non-treated subjects after six months (Engebretson et al., 2013). Thus, to be able to translate the results from systematic reviews and meta-analyses into guidelines for general practice, factors that explain the large differences between individual trials need to be addressed. First, if periodontitis is causally related to poorer metabolic control in DM patients, the amount of periodontal inflammation that is present and removed by treatment, might influence the magnitude of reduction in HbA1c levels. So far, there is no international consensus how to quantify the amount of periodontal inflammation or how to measure the effect of reduction in periodontal inflammation on metabolic control. Critically evaluating the present clinical trials (Chen et al., 2012, Engebretson et al., 2013, Jones et al., 2007, Katagiri et al., 2009, Kiran et al., 2005, Koromantzos et al., 2012, Moeintaghavi et al., 2012, Promsudthi et al., 2005, Singh et al., 2008, Stewart et al., 2001, Sun et al., 2010, Sun et al., 2011, Telgi et al., 2013), patients with various severities of periodontitis (mild, moderate and/or severe periodontitis) are included and the periodontal treatment is often different, varying from initial treatment with (Jones et al., 2007, Katagiri et al., 2009, Promsudthi et al., 2005,
Singh et al., 2008, Sun et al., 2010, Sun et al., 2011) or without (Chen et al., 2012, Engebretson et al., 2013, Kiran et al., 2005, Koromantzos et al., 2012, Moeintaghavi et al., 2012, Singh et al., 2008, Stewart et al., 2001, Telgi et al., 2013) supportive antibiotics, with (Sun et al., 2010, Sun et al., 2011) or without (Chen et al., 2012, Engebretson et al., 2013, Jones et al., 2007, Katagiri et al., 2009, Kiran et al., 2005, Koromantzos et al., 2012, Moeintaghavi et al., 2012, Promsudthi et al., 2005, Singh et al., 2008, Stewart et al., 2001, Telgi et al., 2013) an additional surgical phase, and with (Koromantzos et al., 2012, Moeintaghavi et al., 2012, Stewart et al., 2001, Sun et al., 2010, Sun et al., 2011) or without (Chen et al., 2012, Engebretson et al., 2013, Jones et al., 2007, Katagiri et al., 2009, Kiran et al., 2005, Promsudthi et al., 2005, Singh et al., 2008, Telgi et al., 2013) extractions of hopeless teeth. Most of the treatment regimens were unsuccessful in solving all periodontal problems, resulting in subjects with residual periodontal inflammation at the end of the study period. Second, since metabolic dysregulation negatively affects the periodontal treatment outcome (Lalla and Papapanou, 2011, Llambes et al., 2015), the metabolic control of included subjects at baseline is important.

The metabolic regulation of subjects included in the present studies ranges from well controlled (HbA1c <7%) to poorly uncontrolled (HbA1c >9.5%). So far, the influence of variations in metabolic control on the clinical effects of periodontal treatment and back on metabolic control has not been studied. Third, the various studies include patients with varying levels of obesity and co-morbidities. These factors influence metabolic control and may affect favorable effects of periodontal therapy in a negative way. Fourth, the interaction between two complex diseases has been studied, meaning that several other patient-related factors, such as genetics, lifestyle and biofilm composition might directly or indirectly influence the metabolic control or periodontal treatment outcome or both.

None of these factors have been studied together in relation to periodontal treatment and the effect on metabolic control.

To date, the current (Chapter 3) and newer reviews (Engebretson and Kocher, 2013, Mauri-Obradors et al., 2015, Sgolastra et al., 2013, Simpson et al., 2015, Teshome and Yitayeh, 2016) indicate positive effects of periodontal treatment on metabolic control in type 2 DM patients. However, these studies do not completely prove a causal relationship, i.e. that periodontitis is also involved in the causal chain of events in the initiation and progression of type 2 DM. Nevertheless, all studies so far show that reducing periodontal inflammation apparently helps to improve metabolic regulation in type 2 DM patients (Figure 2). However, the systematic reviews and meta-analyses describing the effect of treating a complex disease on the course of another complex disease is existent, but the outcomes need to be interpreted with the above-mentioned shortcomings in mind.
Figure 2 The relationship between periodontitis, diabetes mellitus and atherosclerotic cardiovascular disease, depicted in outlines of the Bermuda Triangle. The studies in this thesis are mainly focused on the relationship between periodontitis and diabetes mellitus (black-blue line) and periodontitis and atherosclerotic cardiovascular disease (black-red line). The results suggest an associative (straight line) and/or a causative (arrow) relationship. In this perspective, some studies also indirectly discuss other existent relationships (light-colored arrow).

Implementation of oral care in diabetes care
In general, standard diabetes care is focused on improving metabolic control, thereby reducing insulin resistance, and thus prevention of complications (American Diabetes Association, 2015, Rutten et al., 2013). In the Netherlands, diabetes patients are regularly checked for common diabetes complications in a preventive manner, especially for retinopathy, neuropathy and nephropathy (Rutten et al., 2013). Because of the above-mentioned bi-directional relationship between DM and periodontitis, implementation of oral care in diabetes care is desirable. Therefore, a first attempt has been made to
investigate the possibility for internists to obtain current oral health information from the dentist to complete the health assessment for their diabetes patients (Chapter 4). A total of 889 DM patients were available. An oral health questionnaire (OHQ) for professional use was developed. A chain was established from the internist to the patient and from the patient to the dentist, and vice versa, or direct from the dentist to the internist. However, a low response rate (<50%) for information on oral health of their patients with DM was observed. The internists clearly were not used to incorporate questions about oral health into their practice, and vice versa not all dentists were willing to fill out the OHQ. Nevertheless, evaluation of the returned OHQs suggested that the OHQ could be a valuable tool for identification of diabetes patients with poor oral health, especially untreated periodontal disease. Indeed, as shown before (Chapter 3), proper treatment of periodontitis can be helpful for diabetes management. However, the transfer of information from the dentist to the internist is far from optimal and needs improvement. Clearly, many factors might affect the mutual cooperative attitude of internists and dentists, and as such need further investigation in prospective studies.

PERIODONTITIS AND ATHEROSCLEROTIC CARDIOVASCULAR DISEASE

Several epidemiological studies demonstrated an independent association between periodontitis and atherosclerotic cardiovascular disease (ACVD) (Lockhart et al., 2012). Recently, also in the Netherlands, an association between periodontitis and ACVD has been shown (Beukers et al., 2016). ACVD is a collective name for vascular pathologies in which the process of atherogenesis is the underlying etiologic mechanism. A possible explanation for the observed association between periodontitis and ACVD might be the overlapping genetics (i.e. ANRIL, plasminogen) (Schaefer et al., 2015, Schaefer et al., 2013, Schaefer et al., 2011) and lifestyle (i.e. smoking, overweight) (Suvan et al., 2011, Warnakulasuriya et al., 2010) risk factors for both diseases. Next to these risk factors between ACVD and periodontitis, a direct effect of periodontitis on ACVD has been suggested and might also explain the observed association between both diseases. Chronic metabolic or inflammatory diseases, such as DM, rheumatoid arthritis and periodontitis, have been proposed to cause a pro-inflammatory state and may enhance atherogenesis (Kammoun et al., 2014, Kraakman et al., 2016, Schenkein and Loos, 2013). Several causal mechanisms have been proposed, whereby bacterial pathogens, antigens, endotoxins, and/or inflammatory cytokines from the periodontal lesions contribute to the process of atherogenesis and/or to thromboembolic events, thereby increasing the risk for ACVD (Lockhart et al., 2012, Schenkein and Loos, 2013, Tonetti et al., 2013). Periodontal therapy reduces periodontal inflammation and several treatment studies are highly suggestive that this reduction results in lower levels of markers of systemic inflammation (Bokhari et al., 2012, Sun et al., 2011) and improved vascular health (D’Aiuto et al., 2013). In this perspective, C-reactive protein (CRP), an acute phase reactant, is often used as a
biomarker for systemic inflammation. Thereby, CRP is a widely accepted key marker of atherosclerosis and is strongly associated with an increased risk for ACVD (Geluk et al., 2008, Ridker et al., 2010). On average periodontitis patients show elevated C-reactive protein levels compared to controls (Paraskevas et al., 2008) and several studies indicated that periodontal treatment might reduce CRP-levels (D’Aiuto et al., 2013), suggesting that periodontitis causes an increased pro-inflammatory state and contributes to atherosclerosis and an increased risk for ACVD.

To achieve better understanding of the association between periodontitis and ACVD, a study was performed to explore whether genetic variation could explain the observed increased systemic inflammation in periodontitis patients to some extent (Chapter 5). Recently, ANRIL, a genetic risk factor for several conditions with inflammatory components in Caucasians, and the strongest susceptibility locus for several types of ACVD, like coronary artery disease (CAD), myocardial infarction, abdominal aortic aneurysm and intracranial aneurysm (Deloukas et al., 2013, Helgadottir et al., 2008, Schunkert et al., 2008) has been associated with periodontitis (Schaefer et al., 2013, Schaefer et al., 2011). Furthermore, it has been speculated that genetic variation in ANRIL may modulate inflammatory processes (Bochenek et al., 2013, Cunnington et al., 2010). Therefore, the association between levels of CRP and the leading ACVD- and periodontitis-associated ANRIL gene single nucleotide polymorphism (SNP) was investigated across controls and periodontitis patients. From 171 healthy subjects from North European descent (115 periodontitis and 56 controls), CRP levels were determined and subjects were genotyped for the ANRIL SNP (rs1333048). In a multivariate analysis, periodontitis, female gender, increasing BMI and homozygosity for the major allele (AA-genotype) of the ANRIL SNP were significantly associated with elevated CRP plasma levels. This study is the first to show that, in addition to gender and BMI, also a leading SNP in ANRIL is explanatory for inter-individual variation in CRP levels in periodontitis patients of North European descent.

Since CRP levels exhibit a continuous association with the risk of coronary heart disease, ischemic stroke and vascular mortality (Kaptoge et al., 2010), these observations suggest that the complex relationship between periodontitis and ACVD can also be modulated by genetic variation in ANRIL (Figure 2). Interestingly, previous studies showed that carrierhip of the minor allele (C-allele) of ANRIL is associated with ACVD and periodontitis (Deloukas et al., 2013, Helgadottir et al., 2008, Schaefer et al., 2013, Schaefer et al., 2011, Schaefer et al., 2009, Schunkert et al., 2008). In this study (Chapter 5), these findings were corroborated; the minor allele was more frequent in periodontitis patients than controls. Since chronic elevated CRP levels are considered as a prognostic risk marker for ACVD (Geluk et al., 2008, Ridker et al., 2010), an association between the minor allele of ANRIL and increased levels of CRP was expected. However, it was shown here, that, in the
inflammatory disease periodontitis, carriage of the minor allele was associated with lower plasma levels of CRP compared with those that were homozygous for the major allele (AA-genotype). A possible explanation might be that, although chronic elevated CRP levels are considered as a prognostic risk marker for ACVD (Geluk et al., 2008, Ridker et al., 2010), in the initial phase of inflammation, due to a bacterial challenge, CRP might exert a protective role (Du Clos, 2013). When opsonized to a surface, for instance a bacterium, it can activate the classical pathway of the complement system (Holers, 2008, Mihlan et al., 2011, Mold et al., 1984, Suankratay et al., 1998). In that sense, genetic variations inhibiting a rise in CRP, might cause an increased susceptibility for chronic diseases, such as periodontitis and ACVD. Because the current results are the first to suggest this explanation, replication studies in other Caucasian and non-Caucasian periodontitis patients, as well as biochemical studies are necessary to explain the role of ANRIL as pleiotropic genetic locus affecting inflammatory pathways of different immune-mediated diseases (Parkes et al., 2013), like periodontitis and ACVD.

To obtain further understanding of the association between periodontitis and the cardiovascular condition, a study was initiated to investigate the relationship between subclinical atherosclerosis and periodontitis using arterial stiffness (AS) as a recently identified parameter for atherosclerosis (Liu et al., 2011) and ACVD (Greenland et al., 2010) (Chapter 6). Pulse-wave velocity (PWV), a non-invasive chair-side function test for AS, was measured in 57 periodontitis patients, without co-morbidity, and compared to 48 subjects from a reference group. Periodontitis patients showed a significantly increased PWV compared to the reference group and this increase remained significant after adjustments for ACVD risk factors; it can be concluded that periodontitis is associated with increased AS. This confirms with another and new parameter the association of periodontitis with ACVD (Lockhart et al., 2012) (Figure 2). Many studies have shown that periodontal therapy may improve vascular health (D’Aiuto et al., 2013). Also described in Chapter 6, 45 patients with periodontitis were followed for 6 months after periodontal treatment, to explore a possible beneficial effect on arterial function. After periodontal therapy, a reduction in AS (measured by PWV) was seen, but this failed significance. Notably, systolic blood pressure was significantly reduced after periodontal therapy, indicating an improved vascular health, despite the fact that this was not seen for AS. Meanwhile, Vidal and co-workers showed a significantly reduced AS after periodontal treatment in periodontitis patients with refractory hypertension (Vidal et al., 2013), suggesting that especially subjects with co-morbidities, such as hypertension, benefit the most from periodontal treatment.

Periodontal treatment and the atherosclerotic profile
To further explore the beneficial effects of periodontal treatment on the ACVD risk profile, a systematic review and meta-analyses were designed (Chapter 7). From 3928 screened
studies, using three online databases, 25 trials met the eligibility criteria: randomized controlled trials (RCT) or clinical controlled trials (CCT) with periodontitis patients, in which the intervention group received periodontal therapy and the non-intervention group received no periodontal treatment. These trials enrolled 1748 periodontitis patients. Seven trials enrolled periodontitis patients that were otherwise healthy, 18 trials recruited periodontal patients with various co-morbidities, such as ACVD or DM. None of the included trials used hard clinical endpoints of ACVD (i.e. clinical event, such as angina pectoris, myocardial infarction, stroke, death), but only serum/plasma biomarkers or surrogate clinical parameters for atherosclerotic disease (e.g. intima media thickness, flow-mediated dilation, PWV). In fact, these latter clinical parameters reflect endothelial function, a measure of the condition of the vascular system. Although assessed with different methods, all trials, that studied endothelial function, showed improvement of endothelial function after periodontal treatment (Higashi et al., 2009, Higashi et al., 2008, Li et al., 2011, Tonetti et al., 2007). Moreover, the meta-analyses, presented in Chapter 7 demonstrated significant improvement in levels of biomarkers for ACVD, such as CRP, interleukin-6, tumor necrosis factor-α, fibrinogen, triglycerides, total cholesterol, high density lipoprotein cholesterol and HbA1c, all favoring periodontal therapy.

Despite the above-mentioned positive effects, large differences between individual controlled trials have been observed. Similar factors, like the amount of periodontal inflammation at baseline, the treatment regime of periodontitis and patient characteristics, as discussed in the other systematic review (Chapter 3), might explain the observed heterogeneity. Therefore, sub-analyses including co-morbidity, smoking habits and overweight have been performed, since these factors affect ACVD risk (Perk et al., 2012), the periodontal treatment outcome (Lakkis et al., 2012, Warnakulasuriya et al., 2010) or both. These sub-analyses show that for the majority of biomarkers for atherosclerosis, a larger reduction after periodontal treatment could be observed in patients with co-morbidity compared to otherwise healthy patients. In addition, periodontitis patients without overweight or without smoking habits benefitted the most from periodontal therapy, regarding the reduction in levels of biomarkers for atherosclerosis. From the systematic review in Chapter 7 and corresponding meta-analyses, it can be concluded, that periodontal treatment improves endothelial function and reduces biomarkers of atherosclerotic disease (Figure 2), especially in those patients already suffering from ACVD and/or DM. Notably, risk factors for ACVD and DM, like overweight and smoking habits may frustrate the favorable effect of periodontal treatment on the investigated parameters.

**CRP in the oral cavity**

As discussed above, CRP is a widely used biomarker for systemic inflammation and increased risk for ACVD (Geluk et al., 2008, Ridker et al., 2010). In periodontitis, CRP levels
in plasma and saliva correlate with the severity of periodontitis and after periodontal treatment, CRP levels in both plasma and crevicular fluid decrease (Bertl et al., 2013, D’Aiuto et al., 2013, Paraskevas et al., 2008, Torumtay et al., 2016). However, the actual functional role of CRP in periodontitis and maintenance of oral health is unknown. It has been shown that bacterium-bound CRP functions as opsonin, which enhances recognition of bacteria by neutrophils and monocytes, and increases efficacy of phagocytosis as well as cytokine production (Du Clos and Mold, 2004). Next to the possibility to activate the complement cascade via the classical pathway, it has been shown that CRP can inhibit the alternative complement pathway by recruiting factor H (Holers, 2008, Mihlan et al., 2011, Mold et al., 1984, Suankratay et al., 1998). Factor H belongs to the group of complement regulatory proteins and inhibits the alternative pathway amplification loop and the C5 convertase. In particular the latter is required for the formation of the membrane attack complex, which can induce tissue damage (Du Clos and Mold, 2004). In this scenario, the complement cascade is activated to enhance the clearance of invading bacteria, whereas at the same time its deleterious effects on host tissues remain limited (Du Clos, 2013). Therefore, it has been suggested that in the initial phase of inflammation, due to bacterial challenge, the role of CRP might be protective (Du Clos, 2013).

In addition to CRP, it is known that salivary agglutinin (SAG) is a potential activator of the complement cascade via the lectin pathway (Leito et al., 2011, Reichhardt et al., 2012). It has been suggested that when SAG is bound to a bacterial surface, complement proteins may bind to SAG and activate downstream complement pathways, while SAG in solution might prevent complement proteins from binding to other surfaces, thereby inhibiting further activation and subsequent inflammation (Leito et al., 2011, Reichhardt et al., 2012).

Since the aberrant inflammatory response to the dental biofilm is suggested to be the major factor determining the onset and/or progression of periodontitis (Loos et al., 2015), all aspects of immune reactions need consideration. It has been postulated that the complement system is an essential initiator of the inflammatory response and plays an important role in the immune reactions in periodontitis (Hajishengallis et al., 2015). The complement system can be activated via three different pathways: the classical pathway, the lectin pathway and the alternative pathway. In particular, the classical pathway and lectin pathway have a critical role in pathogen recognition and initiation of the complement cascade (Merle et al., 2015). In this perspective, CRP and SAG may play a role in maintaining oral health as well as in periodontitis by initiating the complement pathway. Therefore the hypothesis was put forward that if CRP and SAG participate in maintaining oral health and have a role in periodontitis by modulating the innate immune responses to oral bacteria, CRP and SAG should be present in different ecological niches of the oral cavity. An explorative study (Chapter 8) was performed to investigate the
presence of CRP and SAG in the oral cavity in healthy and inflammatory (i.e. periodontitis) conditions. Subsequently, since both the classical and lectin pathway lead to further downstream activation of the complement system via complement protein C4, the presence of C4 was also determined. Therefore, the presence of CRP, SAG and C4 was determined in supragingival and subgingival plaque, saliva, and salivary cell fraction of 10 healthy subjects and 10 periodontitis patients and in saliva and salivary cell fraction of 10 edentulous subjects. The results showed that CRP, SAG and C4 were present in dental plaque and in saliva of the oral cavity of controls, periodontitis patients and edentulous subjects.

Knowing that CRP and SAG can modulate the innate immune responses to oral bacteria and demonstrating the presence of CRP, SAG and C4 in supra- and subgingival plaque, saliva and salivary cell fraction in healthy and inflammatory conditions, it can be suggested that these proteins play a role in maintaining oral health and are part of various pathways in the pathophysiology of periodontitis.

CONCLUSION

The research presented in this thesis contributes to the understanding of the complex relationship between periodontitis and DM, and periodontitis and ACVD (Figure 2). It was observed that a substantial number of suspected new DM patients could be found in patients with periodontitis. Furthermore, periodontitis patients showed increased CRP levels and more AS compared to controls, reflecting an increased atherosclerotic profile and risk for ACVD. Moreover, periodontal treatment improves the levels of molecular and clinical biomarkers for metabolic regulation and atherosclerosis. However, patient characteristics, such as genetics, lifestyle and the presence of co-morbidities contribute to the complexity of this relationship. Based on these observations and in agreement with a recent consensus report of the joint EFP/AAP workshop on periodontitis and systemic diseases, it is recommend that cardiologists, diabetologists and family physicians ask their patients to be screened by dental care professionals for the presence of periodontitis. If so, these patients should undergo periodontal therapy, which helps to improve their metabolic status and lower their cardiovascular risk profile. This again may reduce the risk for future occurrence of ACVD and DM related complications. In addition, it is proposed that periodontists should consider that their patients with severe periodontitis may have DM. Referral to the family physician or perhaps future ‘point of care’ testing devices may help with this. When a new case of DM is identified, treatment to improve metabolic status can be initiated. Reciprocally, this will contribute to a successful treatment of periodontitis.
REFFERENCES


Chapter 9


