Association of Periodontal Destruction and Diabetes with Mortality

Kebede, T.G.; Holtfreter, Birte; Kocher, Thomas; Meisel, P; Dietrich, Thomas; Biffar, R; Dorr, M; Völzke, Henry; Pink, C.

Document Version
Peer reviewed version

The Association of Periodontal Destruction and Diabetes with Mortality

T.G. Kebede[1] B. Holtfreter[1] T. Kocher[1] P. Meisel[1] T. Dietrich[2] R. Biffar[3] M. Dörr[4,5] H. Völzke[6]
C. Pink[1]

[1]Unit of Periodontology, Department of Restorative Dentistry, Periodontology, Endodontology, Preventive and Pediatric Dentistry, University Medicine Greifswald, Greifswald, Germany

[2]Department of Oral Surgery, School of Dentistry, University of Birmingham, Birmingham, UK

[3]Department of Prosthodontics, Gerodontolgy and Biomaterials, University Medicine Greifswald, Greifswald, Germany

[4]DZHK (German Center for Cardiovascular Research), Partner site Greifswald, Greifswald, Germany

[5]Department of Internal Medicine B, University Medicine Greifswald, Greifswald, Germany

[6]Institute for Community Medicine, Ernst-Moritz-Arndt-University Greifswald, Greifswald, Germany

Corresponding author:

T.G. Kebede, Unit of Periodontology, Department of Restorative Dentistry, Periodontology, Endodontology, Preventive and Pediatric Dentistry, University Medicine Greifswald, Rotgerberstr. 8, 17475 Greifswald, Germany.

Email: getachewkt@uni-greifswald.de

Abstract

Current evidence indicates effects of periodontitis on diabetes as well as on mortality for which diabetes itself represents a risk factor. However, possible interaction of these two chronic conditions regarding mortality has not yet been investigated. The purpose of this study, therefore, was to evaluate whether periodontal destruction interacts with diabetes on all-cause and cardiovascular disease (CVD) mortality or if diabetes serves as a mediator in this association. The study sample comprised 3327 participants from the Study of Health in Pomerania aged 20-81 years. Periodontal destruction was assessed via clinical attachment level (CAL) and the number of missing teeth. Information on mortality (date and ICD10-code) was ascertained from death certificates. Directed acyclic graphs were used to identify potential confounders and Cox proportional hazard models were applied. In 36,701 person-years of follow-up, 263 study participants deceased, thereof 89 due to CVD. Fully adjusted main effect models resulted in hazard ratios of 1.01 (95% CI: 1.002; 1.01) for extent CAL$\geq$3mm, 1.10 (1.03; 1.18) for mean CAL, and 1.03 (1.01; 1.04) for the number of missing teeth regarding all-cause mortality. Analogous results were obtained for CVD mortality with hazard ratios of 1.01 (0.99; 1.02), 1.10 (0.98; 1.23), and 1.02 (0.99; 1.05) for extent CAL, mean CAL and the number of missing teeth, respectively. Findings did not indicate additive interaction of periodontal destruction and diabetes regarding all-cause and CVD mortality. Similarly, no substantial evidence was found to demonstrate the presence of multiplicative interaction or mediation. Besides adjustment for baseline covariates, time-varying covariates were also considered and led to comparable results. In summary, despite their reciprocal relationship, periodontal destruction and diabetes may be independent risk factors for all-cause and CVD mortality.

Introduction

Periodontitis is a chronic inflammation of the tooth-supporting structures. It can result in an irreversible loss of attachment and alveolar bone which may ultimately cause tooth loss. Periodontitis represents a major oral infection contributing to the global burden of chronic diseases (Kassebaum et al., 2014) and according to the World Health Organization, the worldwide prevalence of deep periodontal pockets (≥6 mm) in adults amounts to 10-15% (Petersen and Ogawa, 2012).

Recent literature considers periodontitis as a systemic condition rather than a local inflammation (Hein, 2009; Linden et al., 2013) and a possible link between periodontitis, CVD and thereby mortality has been intensively discussed. Moreover, extensive research was done on the suggested bidirectional relationship of periodontitis and diabetes (Taylor et al., 2013). In particular, many of the latest studies emphasize the impact of periodontitis on glycemic control and diabetes (Demmer et al., 2010; Hasturk and Kantarci, 2015; Taylor et al., 2013). Even though periodontitis and diabetes are two independent conditions, they share a common trait of illness, namely chronic inflammation (Hasturk and Kantarci, 2015). While the biological mechanism by which periodontitis impacts diabetes is not entirely known, up-regulation of proinflammatory cytokines are regarded as the central pathogenic factor in diabetes (Kolb and Mandrup-Poulsen, 2010). The entry of periodontal pathogens into the bloodstream which enhances systemic inflammation is considered as a plausible mechanistic explanation for the impact of periodontitis on diabetes (Chapple and Genco, 2013).

Diabetic subjects have an excess mortality risk mainly due to CVD as compared to non-diabetic subjects (Jansson et al., 2010) and about every second diabetes patient finally dies because of CVD (Morgan et al., 2000). Moreover, because diabetes as well as periodontitis is both considered as having an effect on CVD and mortality via systemic inflammatory processes, the possibility of interaction or mediation due to common pathways was raised (Demmer et al., 2008; Saremi et al., 2005; Southerland et al., 2012). But so far, there is little knowledge on the interplay of periodontal destruction and diabetes regarding mortality. Thus, based on longitudinal data from the Study of Health in Pomerania (SHIP), we investigate how periodontal destruction and diabetes act together in relation to all-cause and CVD mortality and whether diabetes may represent a mediator for the effects of periodontal disease.

Material and Methods

Study population

SHIP is a longitudinal population-based health survey in West Pomerania, a region in northeast Germany (John et al., 2001). The total population in West Pomerania comprised 212,157 inhabitants. A two-stage cluster sampling was adopted from the World Health Organization (WHO) Monitoring of Trends and Determinants in Cardiovascular Disease (MONICA) Project, Augsburg, Germany (Keil et al., 1998). Accordingly, Caucasian subjects with German citizenship and main residency in the study area were randomly sampled within twelve 10-year age strata for both genders, each including 292 subjects. The remaining sample (excluding emigrated or deceased) comprised 6265 eligible subjects of which 4308 finally participated (response rate 68.8%). The study protocol was approved a priori by the local Ethics Committee of the University of Greifswald and written informed consent was obtained from each participant.

Baseline examinations were performed in 1997-2001 and comprised computer-assisted health-related interviews, dental and medical examinations, and risk factor-related questionnaires. Out of 4308 individuals (2116 men), 499 were edentulous. An oral examination was not available in 20 participants. Further, clinical attachment level (CAL) was not assessed in 222 persons because all teeth were crowned. Due to missing data on covariates (N=207), and exclusion of individuals with self-reported cancer (N=32), 3327 participants with complete data were left for final analyses (Supplemental Figure1). All participants gave written informed consent and the study protocol was approved a priori by the local Ethics Committee.

Mortality follow-up

Information on vital status was collected at annual intervals from study enrolment date to September 20, 2011. Subjects got censored at death, failure to follow-up or end of follow-up. The number of months between recruitment and censor date was used as follow-up period. The mean duration of follow-up was 11 (SD =1.73) years. Death certificates were requested from the local health authority at the place of death and were coded by a certified nosologist according to the International Classification of Diseases, 10th revision (ICD-10). Additionally, two internists independently validated the underlying cause of death and performed a joint reading together with a third internist in cases of disagreement.

Periodontal status

Periodontal measurements were obtained at four sites (mesiobuccal, mid-buccal, distobuccal, and mid-lingual/mid-palatal) per tooth according to the half-mouth method, alternating on the left or right side, excluding third molars. A periodontal probe was used (PCP-11, Hu-Friedy, Chicago, IL, USA). Periodontal examinations comprised assessment of probing depth (PD) and CAL. PD was measured as the distance between free gingival margin and cementoenamel junction. CAL equals the distance between the cementoenamel junction and the pocket base. Where the determination of the cementoenamel junction was indistinct (e.g. wedge-shaped defects, fillings, or crown margins), CAL was not recorded. Measurements were mathematically rounded to the nearest millimeter. Periodontitis was assessed as the percentage of sites (extent) with CAL$\geq$3mm and mean CAL on subject level. The number of missing teeth was counted excluding third molars.

Selection of covariates

To model the putative association between exposure, outcome, and covariates in the present study, directed acyclic graphs (DAGs) were used (Supplemental Figure2). DAGs are used to model the causal structure that is thought to underlie the exposure-outcome association of interest (Akinkugbe et al., 2016) and to thereby minimize bias in confounder selection (Merchant and Pitiphat, 2002). Upon proper specification of the DAG, an optimized set of confounders can be achieved (Weng et al., 2009). Furthermore, DAGs performed equally or superior to conventional selection methods such as change-in-estimate or stepwise regression procedures (Weng et al., 2009). However, they often provide two or more different optimized confounder sets (Schwahn et al., 2013). For the present analyses, we constructed a series of slightly different DAGs (e.g. inclusion/exclusion of arrows with limited evidence) to identify a robust confounder set. Finally, using periodontal destruction as exposure and mortality as the outcome, the minimal sufficient adjustment set for the primary DAG (Supplemental Figure 2) included: age, gender, socio-economic status, obesity, smoking, physical activity and oral health behavior.

Measurement of covariates

Information on age, gender and education (<10, 10 or >10 years of education) were taken from the interview. The monthly income was divided by the square root of the household size (Kawachi and Kennedy, 1997) and categorized into tertiles. Oral health behavior was assessed using the question "How often have you been to the dentist in the last 12 months?" Smoking status was categorized as never, current or former smokers. Study participants were considered as being physically active if they did more than one hour of physical exercise per week over summer or winter. Body Mass Index (BMI) was categorized as normal weight (<25kg/m^2), overweight (25-29.9kg/m^2) and obesity ($\geq$30 kg/m^2) according to the WHO (WHO, 2000) criteria. Diabetes mellitus was defined as self-reported physician's diagnosis, treatment with insulin or anti-diabetic medication (ATCcodeA10) or HbA1c-levels$\geq$6.5% or non-fasting blood glucose level$\geq$11.1mmol/l.

Statistical analyses

Summary statistics of baseline characteristics for the study sample were computed as mean$\pm$ standard deviation (SD) or as number (percentage) (Table1 and Supplemental Table1). Multivariable Cox proportional hazard models were used to evaluate the association between periodontal variables and all-cause mortality. The Proportional hazards assumption was evaluated graphically and with χ^2-tests based on Schoenfeld's residuals. All models were adjusted for the minimal sufficient adjustment set which was derived from DAG analyses.

The standard Cox proportional hazard model is not appropriate for identifying risk factors for the cumulative incidence of specific event in the presence of competing risks (Kim, 2007). Since selection bias may be introduced by excluding subjects with competing events (Hernan et al., 2004), competing risk hazard models based on Fine and Gray (Fine and Gray, 1999) were fitted for CVD mortality analyses. In these models, CVD mortality was set as the primary event of interest and death from another cause was treated as the competing event. Accordingly, sub-hazard ratios (SHRs) were reported.

We additionally ran interaction models to analyze the joint effect of periodontal destruction and diabetes on the risk of all-cause or CVD mortality. Accordingly, interaction effects were reported on the additive and multiplicative scale. Regarding additive interaction, the relative excess risk due to interaction (RERI) was calculated. Besides adjustment for baseline covariates, time-varying covariates were also considered to thoroughly explore the strength of relationships.

Moreover, mediation models were built to analyze whether periodontal destruction has an indirect effect on mortality via diabetes. Following the concepts of Mackinnon et al. and Valeri & Vanderweele (MacKinnon et al., 2007; Valeri and Vanderweele, 2013), we estimated the direct, indirect, and marginal total effects. However, these estimated direct and indirect effects may only be interpreted as causal if certain assumptions (like no unmeasured confounders) hold and models are specified correctly (VanderWeele and Vansteelandt, 2009) .We followed the chronological sequence of exposure, mediator, and outcome by measuring periodontal destruction at SHIP-0 and diabetes at SHIP-1 (5-years later). In doing so, mediation analyses were restricted to subjects who survived longer than SHIP-1 examination (2452 subjects with complete data). All analyses were performed with Stata/MP 12.1 (Stata Corp, College Station, Texas) (StataCorp, 2011). We used the additional Stata module "PARAMED" (Emsley and Liu, 2013) for mediation analyses.

Results

Baseline characteristics

During the mean follow-up period of 11 years (36,701 person-years), a total of 181 men and 82 women died (63 men and 26 women from CVD). The mean age at baseline was 44.5 (SD=14.4) years for survivors and 63.7 (SD=12.2) for non-survivors. As compared to survivors, non-survivors were more often male, less educated, more likely to have diabetes, more often obese, less often physically active, more likely smokers, had a worse periodontal status and had fewer teeth (Table1).

Association between periodontal measures and all-cause mortality

In fully adjusted Cox models, all periodontal measures were associated with increased all-cause mortality. Obtained HRs were 1.01 (95%CI: 1.002;1.01) for extent CAL$\geq$3mm, 1.10 (1.03;1.18) for mean CAL, and 1.03 (1.01;1.04) for missing teeth (Table2, upper panel). Regarding interaction analysis (Table2, middle panel), no evidence was observed for a multiplicative interaction of periodontal destruction and diabetes (HRs$\approx$1, p$\geq$0.76). For additive interaction, RERIs were consistent regarding their algebraic sign (all positive) but very small concerning their absolute value (extent CAL: 0.01; mean CAL: 0.08; missing teeth: 0.03). Indirect effects of periodontal destruction on all-cause mortality could not be observed (RRs=1, p$\geq$0.50, Table2, lower panel). Time-varying covariate analyses revealed more or less the same results compared to baseline covariates adjustment (mean CAL: 1.11 (1.05;1.18); missing teeth: 1.03 (1.01;1.04); Supplemental Table3, upper panel).

Association between periodontal measures and CVD mortality

In the same manner, competing risk models (Table3, upper panel) showed associations between dental variables and CVD mortality that were similar to those for all-cause mortality (extent CAL: SHR=1.01; mean CAL:1.10; missing teeth:1.02). Interaction analyses (Table3, middle panel) revealed no evidence of an additional increase in CVD mortality. In mediation analyses, as for all-cause mortality, indirect effects of periodontal destruction on CVD mortality via diabetes could not be substantiated, while the respective RRs equaled one (Table3, lower panel). Again, time-varying covariate analyses produced similar results (mean CAL:1.06 (0.95;1.18); missing teeth:1.01 (0.98; 1.05); Supplemental Table4, upper panel).

Discussion

In the present study, we observed associations between different dental variables and all-cause and CVD mortality after accounting for potential confounders. However, clear evidence towards interaction effects between periodontitis and diabetes could not be identified.

The estimated effects of periodontal destruction on all-cause mortality were modest, although consistent for all dental variables. In fully adjusted multivariable Cox models, a 1mm increase in mean CAL was associated with a 10% higher risk of all-cause mortality and a 1% increase in extent of periodontal sites with CAL≥3mm also was associated with a 1% increase in mortality risk. Analogous analyses for CVD mortality showed similar patterns. Assessment of these associations with time-varying covariate analyses led to similar results as baseline covariate adjustment. All observed associations persisted after further adjustment for hypertension and dyslipidemia in both all-cause (Supplemental Table2 and 5) and CVD mortality analyses (data not shown).

Overall, our findings regarding the relationship between periodontal destruction and mortality are in agreement with previous studies (Ajwani et al., 2003; Janket et al., 2014; Soder et al., 2007; Xu and Lu, 2011). But the absence of any convincing evidence of interaction between periodontal variables and diabetes on mortality did not meet our expectations based on their known common inflammatory mechanisms. From a public health perspective, Rothman suggested the usage of the additive scale to assess interaction (Rothman KJ et al., 2008). A positive departure from additivity of effects implies that the number of events attributable to a combination of two hazards is larger than the sum of the numbers of events that would be associated with the individual risk factors separately (Blot and Day,

1979). Since the obtained RERIs from our interaction analyses on the additive scale were close to zero, we observed no convincing evidence for a joint effect beyond the sum of individual effects.

However, these results seem less surprising against the backdrop of findings from two previous studies. In a cross-sectional study of 6048 persons aged 52-74 (Southerland et al., 2012), the effects of diabetes and periodontitis on the risk of different CVD-related endpoints were evaluated. In fact, diabetes patients with severe periodontitis showed significant Odds for having an intima-media thickness (IMT)>1mm (OR=2.2), acoustic shadowing (OR=2.5) or coronary heart disease (OR=2.6) compared to individuals having neither diabetes nor periodontitis. In this analysis, no significant ORs were found for diabetes or periodontitis alone. But Southerland et al. reported that tests for interaction between diabetes and severe periodontitis on elevated IMT, prevalent coronary heart disease or shadowing were non-significant. Since these results seem quite ambivalent, they may not be taken as comprehensive evidence for the presence of interaction. In the second study on this topic, 204 death cases were observed during a median follow-up time of 11 years in a cohort comprising 628 diabetic Pima Indians (Saremi et al., 2005). Individuals having diabetes and severe periodontal disease had a 3.2 times greater risk of cardiorenal mortality compared to those having diabetes with no, mild or moderate periodontitis (Saremi et al., 2005). Since no reference group of Pima Indians without diabetes was included, no conclusions towards an overall interaction between periodontitis and diabetes were possible. Furthermore, interaction terms between periodontitis and the duration of diabetes were non-significant (Saremi et al., 2005). Since there are notable differences in population characteristics and access to medical care between Pima Indians and Caucasian Germans, the comparability of findings is very limited. But it appears that there may be no additional increase in the risk of mortality due to the interaction of periodontitis and diabetes besides the sum of their individual effects.

Concerning the presented results from our mediation analyses (Tables 2, 3), it was surprising that the indirect effects of periodontal destruction on mortality via diabetes were so small. Even without the existence of an interaction regarding mortality, some mediation via diabetes would be expected according to the current consensus in the literature that periodontitis impairs glycemic control. In NHANES 1, baseline periodontitis represented an independent predictor of incident diabetes in a population-based sample of 9,296 subjects aged 25–74 years (Demmer et al., 2008). Moreover, severe periodontitis was significantly associated with prevalent impaired glucose tolerance (OR= 1.93) in a cross-sectional study among 1,165 adults without diabetes (Arora et al., 2014). Using

SHIP data, Demmer et al. analyzed the association between periodontitis and 5-year progression in HbA1c in 2,973 diabetes-free participants (Demmer et al., 2010). They observed an average HbA1c increase of 0.11% for the highest periodontitis category (highest percentage of sites with CAL≥5 mm) compared to 0.02% for the lowest category.

The reason why there was no evidence for mediation in our analyses may be seen in the characteristics of study participants having diabetes. As presented in Table 1, baseline HbA1c levels differed significantly across survivors and non-survivors for the complete cohort, but not among those having diabetes. About 50% of individuals with diabetes had a good metabolic control at baseline irrespective of survival status and the average HbA1c among individuals with diabetes was about 7.2%. Analogously, for higher quartiles of mean CAL, baseline HbA1c levels were increased in the complete sample but stable in diabetics (Supplemental Table1). During the 5-year follow-up (SHIP-1), individuals with prevalent diabetes at baseline were probably treated to a large extent which is why HbA1c levels for the surviving individuals having diabetes were lower at follow-up (N=195, ΔHbA1c:-0.3, Supplemental Table6). Considering study participants with incident diabetes at SHIP-1, those who died have had higher HbA1c at baseline before they were diabetes patients than those who survived (survivors: 5.8, deaths: 6.1), but both groups showed a cosiderable increase in HbA1c until SHIP-1 (survivors: 0.8, deaths: 0.9). These results may suggest that, in the SHIP cohort, most individuals with diabetes are well-controlled regarding their HbA1c. Hence, in the majority of our diabetes patients, the glycemic control may not have been altered by periodontal infection during the follow-up time and therefore prevented diabetes-mediated effects on mortality. Similarly, the number of uncontrolled diabetes, whose metabolic control could have been impaired by periodontitis, may have been too small in our sample and thereby limited statistical power. However, there is evidence which suggests that HbA1c has a J-shaped relationship with mortality risk in diabetes patients (Arnold and Wang, 2014). Hence, results might be different if individuals with more uncontrolled diabetes were investigated. Another possibility would be that the consequences of a periodontitis-related worsening of glycemic control in diabetes individuals may not be inherently strong enough to be verified by statistical analyses or to reach clinical relevance. But further epidemiological studies will be needed to clarify this aspect.

To the best of our knowledge, this study is the first to analyze the additive interaction of periodontal destruction and diabetes on the risk of mortality as well as the possibility of mediation via diabetes. In doing so, important confounders were identified via DAGs and considered in all statistical

models. Moreover, different statistical modeling approaches were performed to assess changes in the association of interest due to model selection. On top of these, the large sample size compared to previous studies and the long follow-up period count among the strengths of this study. But some limitations have to be mentioned as well. Many individuals had to be excluded from analyses due to different reasons which could have led to selection bias (Hernan et al., 2004) and lower power for statistical tests. Similarly, we considered a very hard clinical endpoint (death) which was not likely to occur with high frequency in a population-based sample aged 20-81 years. As a result, the explanatory power is somehow limited, especially when considering CVD mortality as an outcome. In addition, low prevalence of severe periodontal destruction and diabetes could also preclude mediation effects (Fritz et al., 2015). Finally, we could not entirely exclude unexplained variations due to residual confounding, because periodontal status, diabetes and mortality may relate to other unaccounted factors.

To conclude, this study confirms the association of periodontal destruction with all-cause and CVD mortality based on SHIP data. However, despite their well-known reciprocal relationship, no evidence for interaction or mediation of dental variables and diabetes on mortality was observed; their combined impact on the risk of dying may rather equal the sum of their individual effects. But since this study is based on 263 death cases relative to 36,701 person-years in a population-based cohort with good access to medical care, much larger studies and meta-analyses are required to provide a comprehensive overview on the interplay of periodontal destruction and diabetes on mortality endpoints.

Competing interests

The authors declare that they have no competing interests.

Author contributions

T. Kebede, C. Pink, B. Holtfreter, T. Kocher contributed to conception, design, data acquisition, analysis and interpretation and drafted and critically revised the manuscript; T. Dietrich, R. Biffar, M. Dörr, H. Völzke, contributed to design and interpretation and critically reviewed the manuscript; P. Meisel contributed to analysis and interpretation of results. All authors gave final approval and agreed to be accountable for all aspects of the work.

References

Ajwani S, Mattila KJ, Narhi TO, Tilvis RS, Ainamo A (2003). Oral health status, C-reactive protein and mortality--a 10 year follow-up study. *Gerodontology* 20(1)*:*32-40.

Akinkugbe AA, Sharma S, Ohrbach R, Slade GD, Poole C (2016). Directed Acyclic Graphs for Oral Disease Research. *J Dent Res* 95(8)*:*853-859.

Arnold LW, Wang Z (2014). The HbA1c and All-Cause Mortality Relationship in Patients with Type 2 Diabetes is J-Shaped: A Meta-Analysis of Observational Studies. *The Review of Diabetic Studies : RDS* 11(2)*:*138-152.

Arora N, Papapanou PN, Rosenbaum M, Jacobs DR, Jr., Desvarieux M, Demmer RT (2014). Periodontal infection, impaired fasting glucose and impaired glucose tolerance: results from the Continuous National Health and Nutrition Examination Survey 2009-2010. *J Clin Periodontol* 41(7)*:*643-652.

Blot WJ, Day NE (1979). Synergism and interaction: are they equivalent? *Am J Epidemiol* 110(1)*:*99-100.

Chapple IL, Genco R (2013). Diabetes and periodontal diseases: consensus report of the Joint EFP/AAP Workshop on Periodontitis and Systemic Diseases. *J Periodontol* 84(4 Suppl)*:*S106-112.

Demmer RT, Jacobs DR, Jr., Desvarieux M (2008). Periodontal disease and incident type 2 diabetes: results from the First National Health and Nutrition Examination Survey and its epidemiologic follow-up study. *Diabetes Care* 31(7)*:*1373-1379.

Demmer RT, Desvarieux M, Holtfreter B, Jacobs DR, Wallaschofski H, Nauck M *et al.* (2010). Periodontal Status and A1C Change: Longitudinal results from the Study of Health in Pomerania (SHIP). *Diabetes Care* 33(5)*:*1037-1043.

Emsley R, Liu H (2013). PARAMED: Stata module to perform causal mediation anlysis using parametric regression models.

Fine JP, Gray RJ (1999). A Proportional Hazards Model for the Subdistribution of a Competing Risk. *Journal of the American Statistical Association* 94(446)*:*496-509.

Fritz MS, Cox MG, MacKinnon DP (2015). Increasing Statistical Power in Mediation Models Without Increasing Sample Size. *Eval Health Prof* 38(3)*:*343-366.

Hasturk H, Kantarci A (2015). Activation and resolution of periodontal inflammation and its systemic impact. *Periodontol 2000* 69(1)*:*255-273.

Hein C (2009). Translating evidence of oral-systemic relationships into models of interprofessional collaboration. *J Dent Hyg* 83(4)*:*188-189.

Hernan MA, Hernandez-Diaz S, Robins JM (2004). A structural approach to selection bias. *Epidemiology* 15(5)*:*615-625.

Janket SJ, Baird AE, Jones JA, Jackson EA, Surakka M, Tao W *et al.* (2014). Number of teeth, C-reactive protein, fibrinogen and cardiovascular mortality: a 15-year follow-up study in a Finnish cohort. *J Clin Periodontol* 41(2)*:*131-140.

Jansson SP, Andersson DK, Svardsudd K (2010). Mortality trends in subjects with and without diabetes during 33 years of follow-up. *Diabetes Care* 33(3)*:*551-556.

John U, Greiner B, Hensel E, Ludemann J, Piek M, Sauer S *et al.* (2001). Study of Health In Pomerania (SHIP): a health examination survey in east German region: objectives and design. *Soz Praventivmed* 46(3)*:*186-194.

Kassebaum NJ, Bernabe E, Dahiya M, Bhandari B, Murray CJ, Marcenes W (2014). Global burden of severe periodontitis in 1990-2010: a systematic review and meta-regression. *J Dent Res* 93(11)*:*1045-1053.

Kawachi I, Kennedy BP (1997). The relationship of income inequality to mortality: does the choice of indicator matter? *Soc Sci Med* 45(7)*:*1121-1127.

Keil U, Liese AD, Hense HW, Filipiak B, Doring A, Stieber J *et al.* (1998). Classical risk factors and their impact on incident non-fatal and fatal myocardial infarction and all-cause mortality in southern Germany. Results from the MONICA Augsburg cohort study 1984-1992. Monitoring Trends and Determinants in Cardiovascular Diseases. *Eur Heart J* 19(8)*:*1197-1207.

Kim HT (2007). Cumulative incidence in competing risks data and competing risks regression analysis. *Clin Cancer Res* 13(2 Pt 1)*:*559-565.

Kolb H, Mandrup-Poulsen T (2010). The global diabetes epidemic as a consequence of lifestyle-induced low-grade inflammation. *Diabetologia* 53(1)*:*10-20.

Linden GJ, Lyons A, Scannapieco FA (2013). Periodontal systemic associations: review of the evidence. *Journal of Clinical Periodontology* 40(S8-S19.

MacKinnon DP, Fairchild AJ, Fritz MS (2007). Mediation analysis. *Annu Rev Psychol* 58(593-614.

Merchant AT, Pitiphat W (2002). Directed acyclic graphs (DAGs): an aid to assess confounding in dental research. *Community Dent Oral Epidemiol* 30(6)*:*399-404.

Morgan CL, Currie CJ, Peters JR (2000). Relationship between diabetes and mortality: a population study using record linkage. *Diabetes Care* 23(8)*:*1103-1107.

Petersen PE, Ogawa H (2012). The global burden of periodontal disease: towards integration with chronic disease prevention and control. *Periodontol 2000* 60(1)*:*15-39.

Rothman KJ, Greenland S, Lash TL (2008). Modern Epidemiology 3rd ed.: Lippincott Williams & Wikins.

Saremi A, Nelson RG, Tulloch-Reid M, Hanson RL, Sievers ML, Taylor GW *et al.* (2005). Periodontal disease and mortality in type 2 diabetes. *Diabetes Care* 28(1)*:*27-32.

Schwahn C, Polzer I, Haring R, Dorr M, Wallaschofski H, Kocher T *et al.* (2013). Missing, unreplaced teeth and risk of all-cause and cardiovascular mortality. *Int J Cardiol* 167(4)*:*1430-1437.

Soder B, Jin LJ, Klinge B, Soder PO (2007). Periodontitis and premature death: a 16-year longitudinal study in a Swedish urban population. *J Periodontal Res* 42(4)*:*361-366.

Southerland JH, Moss K, Taylor GW, Beck JD, Pankow J, Gangula PR *et al.* (2012). Periodontitis and diabetes associations with measures of atherosclerosis and CHD. *Atherosclerosis* 222(1)*:*196-201.

StataCorp (2011). Stata Statistical Software: Release 12. College Station, TX: StataCorp LP

Taylor JJ, Preshaw PM, Lalla E (2013). A review of the evidence for pathogenic mechanisms that may link periodontitis and diabetes. *J Clin Periodontol* 40 Suppl 14(S113-134.

Valeri L, Vanderweele TJ (2013). Mediation analysis allowing for exposure-mediator interactions and causal interpretation: theoretical assumptions and implementation with SAS and SPSS macros. *Psychol Methods* 18(2)*:*137-150.

VanderWeele T, Vansteelandt S (2009). Conceptual issues concerning mediation, interventions and composition. *Statistics and Its Interface* 2)*:*457-468.

Weng HY, Hsueh YH, Messam LL, Hertz-Picciotto I (2009). Methods of covariate selection: directed acyclic graphs and the change-in-estimate procedure. *Am J Epidemiol* 169(10):*1182-1190.

WHO (2000). Obesity: preventing and managing the global epidemic. Report of a WHO consultation. *World Health Organ Tech Rep Ser* 894(i-xii, 1-253.

Xu F, Lu B (2011). Prospective association of periodontal disease with cardiovascular and all-cause mortality: NHANES III follow-up study. *Atherosclerosis* 218(2):*536-542.

Table 1 Baseline characteristics of study participants according to mortality status (N=3327).

	Survivors	Total Deaths	CVD Deaths
N	3064	263	89
Observation period, years	11.4±0.8	6.9±3.1*	7±2.9 *
Mean CAL, mm	2.4±1.7	4.6±2.2 *	4.7±2.3 *
Extent CAL ≥3 mm, %	43±34.0	79.2±26.0 *	81±23.7 *
Number of missing teeth ‡	6.5±6.4	14.1±7.7 *	15.0±6.9 *
HbA1c, % #	5.3±0.8	5.9±1.1*	6.0±1.1 *
Diabetes mellitus (yes)	197 (6.4)	75 (28.5) *	28 (31.5) *
controlled diabetes	103 (52.3)	39 (52.0) *	14 (50.0) *
uncontrolled diabetes	94 (47.7)	36 (48.0) *	14 (50.0) *
HbA1c, %¶	7.2±1.5	7.1±1.3	7.1±1.2
Age, years	44.5±14.4	63.7±12.2 *	67.0±9.8 *
Sex (male)	1431 (46.7)	181 (68.8) *	63(70.8) *
School education			
<10 years	867 (28.3)	174 (66.3)	63 (70.8)
10 years	1608 (52.5)	54 (20.5)	12 (13.5)
>10 years	589 (19.2)	35 (13.3) *	14 (15.7) *
Equivalised household income			
1st tertile, < 725 €	1049 (34.3)	86 (32.7)	26(29.2)
2nd tertile, 725-1174 €	1006 (32.8)	86 (32.7)	33(37.1)
3rd tertile, >1174 €	1009 (32.9)	91 (34.6)	30(33.7)
Smoking status			
Never smokers	1111 (36.3)	78 (29.7)	32 (36.0)
Former smokers	960 (31.3)	107 (40.7)	37 (41.6)
Current smokers	993 (32.4)	78 (29.7) *	20 (22.5) *
Body Mass Index			
<25 kg/m²	1171 (38.2)	62 (23.6)	16 (18.0)
25- <30 kg/m²	1188 (38.8)	113 (43.0)	32 (36.0)
≥30 kg/m²	705 (23.0)	88 (33.5) *	41 (46.1) *
Hypertension (yes)	1358 (44.3)	216 (82.1) *	80 (89.9) *
Dyslipidemia (yes)	1383 (45.3)	164 (62.4) *	63 (70.8) *
Physical activity (yes)	1452 (47.4)	81 (30.8) *	26 (29.2) *
Dental check-up during the last year (yes)	2762 (90.1)	211 (80.2) *	74 (83.2) *

Data are presented as mean ± SD or number (%). P-values were obtained using independent samples t-tests (continuous variables) and chi-square tests (categorical variables) comparing people who survived against those who died of all causes or CVD. Equivalised household income was computed as net household income divided by the square root of household size. Smoking status was defined as never, former and current smokers. Diabetes mellitus was defined as self-reported physician's diagnosis or antidiabetic treatment (Anatomical Therapeutic Chemical Classification System (ATC) code A10) or non-fasting glucose levels ≥11.1 mmol/l or glycated hemoglobin (HbA1c) concentrations ≥6.5%. Physical activity was defined as at least 1 h of physical exercise per week during summer or winter. Uncontrolled diabetes was defined considering Diabetes patients with HbA1c ≥7%. Hypertension was defined as systolic blood pressure ≥140mmhg or diastolic blood pressure ≥90mmhg or usage of antihypertensive medication. Dyslipidemia was defined as total cholesterol ≥6.2 mmol/l or low-density lipoprotein cholesterol ≥4.1 mmol/l or high-density lipoprotein cholesterol <1.04 mmol/l or usage of statins.

Abbreviations: CVD, cardiovascular diseases; CAL, clinical attachment level; HbA1c, glycated hemoglobin.

*P<0.001.

HbA1c levels for the entire cohort.

¶ HbA1c levels for individuals having diabetes.

Table 2 Cox and mediation models for associations between dental variables, diabetes, and all-cause mortality

		Extent CAL ≥ 3mm		Mean CAL		Number of missing teeth	
		HR (95% CI)	P-value	HR (95% CI)	P-value	HR (95% CI)	P-value
Model 1	Dental variable	1.01 (1.002; 1.01)	0.01	1.10 (1.03; 1.18)	0.002	1.03 (1.01; 1.04)	0.01
	Diabetes	1.85 (1.40; 2.44)	<0.001	1.84 (1.38; 2.43)	<0.001	1.88 (1.42; 2.49)	<0.001
Model 2	Dental variable	1.01 (1.00; 1.014)	0.01	1.11 (1.03; 1.19)	0.01	1.02 (1.00; 1.04)	0.03
	Diabetes	2.16 (0.74; 6.30)	0.16	1.91 (0.95; 3.84)	0.07	1.75 (0.92; 3.36)	0.09
	Multiplicative interaction	1.00 (0.99; 1.01)	0.76	0.99 (0.87; 1.13)	0.90	1.00 (0.97; 1.04)	0.81
		RERI (95% CI)		RERI (95 % CI)		RERI (95 % CI)	
	Additive interaction	0.01 (-0.01; 0.02)	0.43	0.08 (-0.06; 0.23)	0.28	0.03 (-0.01; 0.06)	0.16
		RR (95% CI)		RR (95% CI)		RR (95% CI)	
Model 3	Total	1.01 (1.00; 1.02)	0.03	1.14 (1.02; 1.28)	0.02	1.02 (1.00; 1.06)	0.23
	Indirect	1.00 (0.99; 1.00)	0.60	1.00(0.99: 1.005)	0.50	1.00 (0.99; 1.00)	0.75
	Direct	1.01 (1.00; 1.02)	0.03	1.14 (1.02; 1.28)	0.02	1.02 (0.99; 1.06)	0.23

Abbreviations: HR: Hazard ratio; CI: Confidence interval; CAL: Clinical attachment level; RERI: Relative excess risk due to interaction; RR: Relative risk. Model 1: Main effect model (N=3327; number of people living with diabetes: N=272, number of deaths: N=263); Model 2: model 1+ interaction terms; Model 3: mediation model considering incident diabetes at SHIP 1 (N=2452; incident diabetes: N=104; number of deaths: N=87). All models were adjusted for age, sex, equivalised household income, years of education, Body Mass Index, smoking, physical activity and dental check-up.

Table 3 Competing risk and mediation models for associations between dental variables, diabetes, and CVD mortality

		Extent CAL ≥ 3mm		Mean CAL		Number of missing teeth	
		SHR (95% CI)	P-value	SHR (95% CI)	P-value	SHR (95% CI)	P-value
Model 1	Dental variable	1.01 (0.99; 1.02)	0.37	1.10 (0.98; 1.23)	0.11	1.02 (0.99; 1.05)	0.17
	Diabetes	1.52 (0.94; 2.47)	0.09	1.51 (0.93; 2.43)	0.09	1.55 (0.96; 2.50)	0.08
Model 2	Dental variable	1.01 (1.00; 1.02)	0.23	1.10 (0.97; 1.25)	0.17	1.02 (0.98; 1.05)	0.31
	Diabetes	3.06 (0.44; 21.2)	0.26	1.49 (0.38; 5.86)	0.57	1.26 (0.41; 3.89)	0.68
	Multiplicative interaction	0.99 (0.97; 1.01)	0.47	1.00 (0.78; 1.29)	0.98	1.01 (0.95; 1.08)	0.70
		RERI (95% CI)		RERI (95 % CI)		RERI (95 % CI)	
	Additive interaction	-0.01 (-0.08; 0.06)	0.75	0.05 (-0.16; 0.26)	0.62	0.02 (-0.02; 0.07)	0.36
		RR (95% CI)		RR (95% CI)		RR (95% CI)	
Model 3	Total	0.99 (0.98; 1.01)	0.51	0.93 (0.72; 1.19)	0.55	0.97 (0.91; 1.05)	0.48
	Indirect	1.00 (0.99; 1.00)	0.45	1.00 (0.99; 1.00)	0.67	1.00 (0.99; 1.00)	0.06
	Direct	1.00 (0.98; 1.01)	0.52	0.93 (0.72; 1.19)	0.56	0.98 (0.91; 1.05)	0.48

Abbreviations: SHR: Sub-hazard ratio; CI: Confidence interval; CAL: Clinical attachment level; RERI: Relative excess risk due to interaction; RR: Relative risk. Model 1: Main effect competing risk model (N=3327; people living with diabetes: N=272, CVD deaths: N=89); Model 2: model 1 + interaction terms; Model 3: mediation models considering incident diabetes at SHIP 1 (N=2452; incident diabetes: N=104; CVD deaths: N=25). All models were adjusted for age, sex, equivalised household income, years of education, Body Mass Index, smoking, physical activity and dental check-up.