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Periodontitis is associated with incident chronic liver disease – a population-based cohort study

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Abbreviations: NAFLD, non-alcoholic fatty liver disease; WHO, World Health Organization; CRP, C-reactive protein; GGT, gamma-glutamyltransferase; HOMA-IR, homeostasis model assessment index; EASL, European Association for the Study of the Liver; ICD, International Classification of Diseases; CDP, Center for Disease and Prevention; AAP, American Academy of Periodontology; CAL, clinical attachment loss; PD, probing depth

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ABSTRACT

Background & Aims

Chronic liver disease is a major health concern worldwide and the identification of novel modifiable risk factors may benefit subjects at risk. Few studies have analyzed periodontitis as a risk factor for liver complications. We studied whether periodontitis is associated with incident severe liver disease.

Methods

The study comprised 6165 individuals without baseline liver disease who participated in the Finnish population-based Health 2000 Survey (BRIF8901) during 2000–2001, a nationally representative cohort. Follow-up was until 2013 for liver-related admissions, liver cancer, and mortality from National Hospital Discharge, Finnish Cancer Registry, and from Causes-of-Death Register, Statistics Finland. Mild to moderate periodontitis was defined as ≥1 tooth with periodontal pocket ≥4 mm deep, and advanced periodontitis as ≥5 teeth with such pockets. Multiple confounders were considered.

Results

79 subjects experienced a severe liver event during follow-up. When adjusted for age, sex and number of teeth, hazards ratios by Cox regression regarding incident severe liver disease were, for mild to moderate periodontitis, 2.12 (95% CI 0.98-4.58), and, for advanced periodontitis, 3.69 (95% CI 1.79-7.60). These risk estimates remained stable after additionally adjusting for alcohol use, smoking, metabolic risk, serum gamma-glutamyltransferase, dental-care habits, lifestyle, and socioeconomic status. Periodontal disease-associated liver risk was accentuated among subjects with non-alcoholic fatty liver disease or heavy alcohol use at baseline.

Conclusions

Periodontitis was associated with incident liver disease in the general population independently of various confounders. As a preventable disease, periodontal disease might present a modifiable risk factor for chronic liver disease.

Keywords: periodontitis, chronic liver disease, bacterial translocation, oral infection, mortality

Key point box

- Experimental periodontitis in animal models has been shown to exert liver damage.
- In this longitudinal epidemiologic study, periodontitis was independently associated with the risk of incident severe liver disease in the general population.
- This association was stronger for individuals with preexisting susceptibility for liver disease (non-alcoholic fatty liver disease or heavy alcohol use at baseline).
- Prevention and treatment of periodontitis may be warranted in patients at risk for chronic liver disease.

Introduction

Periodontitis is a bacteria-induced chronic, multifactorial inflammatory disease that causes destruction of tooth-supporting tissues in the gingiva, periodontal ligament, and alveolar bone. Its prevalence among US adults is 45-58% (1). In Finland the prevalence is around 64% (2). Severe disease affects 5-15% of the population (3-6).

Accumulation of bacterial plaque on the tooth surface induces inflammation (gingivitis). When prolonged, this may lead to periodontal pocket formation – a separation of the coronal portion of the gingiva from the root surface – which is characteristic of chronic periodontitis (7). Such inflamed periodontal pockets can harbor 10^8-10^{10} bacteria, and serve as a focus from where bacteria, microbial products and locally produced inflammatory mediators can enter the blood stream and affect distal organs. Epidemiological studies have found associations between periodontitis and systemic disease, including cardiovascular disease,

kidney disease, obesity, and adverse pregnancy outcomes (8-14). Emerging evidence also suggests a link to liver disease (15-17).

Experimental periodontitis in animal models has been shown to induce liver damage (18-20). A
link between periodontitis, oral and gut dysbiosis, and gut permeability has also been described (21). In non-alcoholic fatty liver disease (NAFLD) patients, treatment of periodontitis was associated with a reduction in serum transaminase levels (22). In another study, poor oral health was associated with an accelerated progression of liver cirrhosis (23).

Based on these data, we hypothesized that periodontitis could affect the risk for future liver disease. We analyzed whether an epidemiologic association exists between periodontal disease and incident liver morbidity and mortality in a large population-based cohort.

Material and methods

Baseline data were extracted from the Health 2000 Survey, which was a multidisciplinary epidemiologic survey in Finland that used a regional two-stage stratified cluster sampling procedure to ensure that the sample was representative of the entire Finnish adult population. The Health 2000 Survey was conducted in 2000–2001, coordinated by the National Public Health Institute (now known as the National Institute for Health and Welfare), and the main sample originally comprised 8028 adults aged 30 years and over. Data were collected via structured home or telephone interviews, self-reported questionnaires, clinical measurements, blood tests, and by clinical examination by a physician and a dentist. The Health 2000 Survey protocol is described in detail elsewhere (24,25). The Epidemiology Ethics Committee of the

Helsinki and Uusimaa Hospital Region approved the Health 2000 Survey protocol, and all participants provided signed informed consent.

Clinical oral examination

Baseline data for this study were gathered from clinical oral health examinations including participants who were edentulous or whose periodontal health was determined, and whose follow-up data from registers were available (n=6165). Quality of clinical assessment was assured through parallel measurements by the five field dentists and a reference dentist (2). Inter-reliability between the five dentists for measurements of probing depth was very good (kappa-value 0.83). The methods used for clinical oral examination were based on previous epidemiologic studies (26-28), and the World Health Organization (WHO) protocols (29,30).

A removable dental unit included a dental chair, an air spray, suction and a fiber optic. The field dentists followed detailed instructions and assessments during the examination and data collection. The mouth and teeth were examined using a dental mirror and a WHO periodontal probe in a standardized order; starting from the upper right most distal tooth and ending at the lower right distal tooth. The intraoral examination included registering the amount of plaque on the tooth surface, possible bleeding on probing, and measurements of periodontal probing depth (2). The periodontal probing depth was registered on dentate subjects separately by each tooth (excluding wisdom teeth) on four different surfaces: no periodontal pocket, 4 – 5mm, or ≥ 6mm periodontal pocket. From this data, a separate variable "number of teeth with deepened periodontal pockets ≥4mm" (sum of all periodontal pockets) was calculated for each subject. Number of teeth was assessed by clinical oral examination, calculating the upper and lower teeth together.

Assessment of periodontitis

Occurrence of periodontitis was defined as at least one tooth with a periodontal pocket at least 4 mm deep. Subjects with such teeth were divided into 2 groups by the median number of teeth with ≥4 mm pockets (=5), that is mild to moderate, and advanced periodontitis. Edentulous patients formed a fourth group. Therefore, we analyzed 4 subgroups based on periodontal health: Group A (no periodontitis): dentate subjects with no periodontal pockets (n=1900), Group B (mild to moderate periodontitis): dentate subjects with 1-4 teeth with ≥4 mm deep periodontal pockets (n=1610), Group C (advanced periodontitis): dentate subjects with 5 or more teeth with ≥4 mm deep periodontal pockets (n=1723), and Group D: edentulous subjects (n=932).

Demographic data

Data from interviews or questionnaires included questions about marital status, educational level and employment, tooth-brushing frequency (at least twice daily, once daily or less often), frequency of dental visits (annually, every two years or less frequently), average alcohol use (grams per day), smoking status (never, former, current smoker), exercise habits (frequency of moderate-intensity or high-intensity physical exercise for at least half an hour), daily use of sweets and sugar-sweetened beverages. Laboratory measurements included fasting serum triglycerides, HDL cholesterol, glucose, insulin, C-reactive protein (CRP), and gamma-glutamyltransferase (GGT).

Metabolic syndrome was defined according to the Joint Interim Statement criteria (31). Diabetes was defined either by fasting serum glucose ≥7.0 mmol/L, taking diabetes medication, or a previous diabetes diagnosis. The homeostasis model assessment index (HOMA-IR), calculated based on fasting serum glucose and insulin values, served as an estimate of insulin resistance (32).

Non-alcoholic fatty liver disease (NAFLD) was defined as a fatty liver index >60 together with average alcohol use <30g/day for men and <20g/day for women (33). The fatty liver index is based on BMI, waist circumference, GGT and triglyceride level, and is a validated method, endorsed by the European Association for the Study of the Liver (EASL) guidelines, to detect NAFLD in large-scale epidemiologic studies (specificity 87% at an index >60).

Follow-up data

Follow-up data for hospital admissions were obtained from the National Hospital Discharge Register, which covers all hospitalizations in Finland starting in 1969. Data for cancers were obtained from the Finnish Cancer Registry, and vital status and cause of death data were obtained from Causes-of-Death Register, Statistics Finland, which systematically collects data about the deaths of all Finnish citizens. In Finland, each person who dies is by law assigned a cause of death (in accordance with the International Classification of Diseases, ICD) to the official death certificate, issued by the treating physician based on medical or autopsy evidence, or forensic evidence when necessary; the death codes are then verified by medical experts at the register and recorded according to systematic coding principle. One or several ICD-diagnoses are assigned to each hospitalization at discharge; these diagnosis codes are systematically recorded in the National Hospital Discharge Register. Data collection to all these registries is obligatory and general quality is consistent and complete. Linkage was performed using the unique personal identifiers assigned to all Finnish residents.

Outcome

The primary outcome event of this study was a first hospitalization due to liver disease or liver-related death or a diagnosis of (primary) liver cancer, whichever came first. In line with previous studies (34-36), liver disease was defined as ICD8/9 codes 570–573 and ICD10

codes K70–K77 and C22. Liver-related death was defined as having one of these diagnoses as the underlying cause of death. Patients were followed for deaths and hospitalizations until December 2013 and for cancers until December 2012.

We excluded participants with records showing that one of the study outcomes (n=37) occurred before study baseline. We also examined alternate outcomes, liver admission or cancer (excluding liver-death without prior known liver disease), and non-liver death without prior known liver disease.

Statistical analysis

For comparing groups, we used chi-square, Mann-Whitney, or Kruskall-Wallis tests as appropriate. We tested the impact of periodontal health on the risk of incident liver disease by Cox regression models with various levels of adjustment. Factors included in these models as potential confounders were identified through literature review, and included age, sex, number of teeth, alcohol use, smoking, metabolic syndrome, waist circumference, diabetes, HOMA-IR, GGT, tooth-brushing frequency, use of sweets and sweetened beverages, exercise, marital status, education, and employment. Subgroup analyses were by alcohol use and presence of NAFLD, smoking, number of teeth, and excluding individuals with chronic viral hepatitis at baseline or during follow-up (ICD10: B18, n=16).

The proportional hazard assumptions of the Cox model were tested for each covariate by visually examining the parallelism of stratified survival curves, but no violations were detected. The relationship between number of $\geq$4 mm deep periodontal pockets and incident liver disease was examined using the penalized spline smoothing method (37) adjusted for age, sex and number of teeth (edentulous subjects were excluded). P values <0.05 were

considered statistically significant. Data were analyzed with SPSS version 23 and R software version 3.2.5.

Results

The study comprised 6165 subjects. During follow-up, we observed 79 incident severe liver events (hospitalization due to liver disease, liver cancer or liver death), with the first event occurring at mean 6.6 years (SD 3.8) after baseline. Of liver events, 35 (44%) occurred in Group C (advanced periodontitis). The first liver event was hospitalization in 59 cases, liver cancer in 9, and liver-related death in 11.

There were significant differences among study groups in several demographic factors (Table 1). These differences were mostly fairly small, except for edentulous subjects (Group D), which differed markedly from the other groups (Table 1).

The mean (SD) number of teeth among all subjects was 22.9 (SD 7.8), varying from 22.4 to 23.8 by study groups A to C (Table 2). The mean (SD) number of teeth with ≥4 mm pockets among all subjects was 3.5 (4.5).

Subjects with worse periodontal condition seemed to have more frequent dental examinations compared to those with better periodontal health (Table 2). A significant tendency towards less frequent tooth-brushing frequency and less interdental cleaning was observed in Group C (Table 2). Periodontal status correlated significantly with visible plaque and bleeding on probing (Table 2). In Health 2000 Survey, bleeding on probing was measured on every sextant and dental plaque on three different index teeth but in this current study, only one of

each is shown here. Dental plaque on lower left lingual tooth seemed to have the highest prevalence of plaque deposits.

The number of ≥4 mm deep periodontal pockets as a continuous variable was significantly associated with severe liver events (hazards ratio 1.04, 95% CI 1.002-1.08, P=0.04) by Cox regression analysis adjusted for age, sex, and number of teeth (edentulous subjects excluded) (Figure 1).

In Cox regression analysis adjusted for age, sex, and number of teeth, Group B (mild to moderate periodontitis) and Group C (advanced periodontitis) exhibited, respectively, 2.1-fold and 3.7-fold increased risks for severe liver events compared to Group A (no periodontitis) (Table 3). The corresponding risk for Group D (edentulous subjects) was 1.5-fold. In multivariate models with various levels of adjustment, the hazards ratios for liver events varied for Group B from 2.17 to 2.35, and for Group C from 3.29 to 3.72 (Table 3).

Data on frequency of dental visits was available for 2972 individuals (48%). Periodontal status remained significant after including also this variable as a confounder in the multivariate model 3 in Table 3 (data not shown).

Subgroup analysis

Of subjects, 10% reported heavy alcohol drinking (average alcohol use ≥30 g/day for men and ≥20 g/day for women) and 31% presented with NAFLD (fatty liver index >60 without heavy alcohol use). The impact of periodontitis status on the risk for liver events was noticeably augmented in the subgroups of heavy drinkers or NAFLD (Table 4). The adjusted hazards ratio for Group C (advanced periodontitis) compared to Group A (no periodontitis)

was 6.9 among subjects with NAFLD and 7.8 among heavy drinkers, whereas it was 2.1 among subjects with neither NAFLD nor heavy drinking (Table 4). Periodontitis status was also significant for liver events in current smokers, never smokers and subjects with more than 20 teeth (Supplemental Table 1). Sixteen subjects were diagnosed with chronic viral hepatitis either at baseline or during follow-up; of these, 3 had a liver event. Exclusion of the 16 subjects with viral hepatitis did not change the findings (Supplemental table 1).

Alternate outcomes

Periodontitis status was significant for liver events also after exclusion of liver-deaths where the liver disease was unknown at time of death (Supplemental Table 2). In contrast, periodontitis status did not predict liver-unrelated deaths (Supplemental Table 2).

Discussion

In this nationally representative epidemiologic study we found that periodontitis was independently associated with the risk of incident severe liver disease. Having advanced periodontitis (5 or more teeth with deep periodontal pockets) increased the risk for liver events by 3- to 4-fold compared to no periodontitis. A higher risk for liver disease with more extensive periodontitis supports a direct link between these two conditions. In addition, the fact that periodontitis was non-significant for liver-unrelated mortality means that the increased risk for liver disease observed in our study is not confounded by competing risk issues. Our study hypothesis was thus confirmed.

The association between periodontitis and incident liver disease remained significant and robust after various levels of adjustment for relevant confounders. Confounders included in the analyses were those identified through literature review to be associated with periodontitis and/or liver disease. We were able to adjust for demographic and social factors,

metabolic risk factors, alcohol use, smoking, lifestyle factors and oral hygiene habits. We were able to exclude subjects with baseline clinical liver disease and adjust for baseline GGT. The differences in baseline characteristics between study groups A, B and C were mostly fairly small. Nonetheless, some degree of residual confounding effect cannot be excluded.

Data came from a large, nationally representative cohort, carefully recorded prospectively in a standardized fashion. Completeness of data in follow-up registries and validity of liver diagnoses are considered high. Study outcomes represented clinically relevant liver disease based on standard diagnosis codes (34-36).

We defined periodontitis as having at least one tooth with at least 4 mm deep periodontal pocket. According to Center for Disease and Prevention (CDP) and the American Academy of Periodontology (AAP) moderate periodontitis is defined by two or more teeth with clinical attachment loss $\geq$ 4mm at interproximal sites, or two or more teeth with pocket probing depth $\geq$5mm at interproximal sites. This classification could not be used in the present epidemiologic survey-based study. Especially before year 2000, there has been several definitions and classifications of periodontitis in previous periodontal research. According to AAP, apart from gingival bleeding and radiographics, basic measures for periodontitis are clinical attachment loss (CAL) and probing depth (PD) but the measurements have varied and there has been no consensus on these matters (38).

A link between periodontitis and liver disease is best described in animal models (18,20,22,39,40), but clinical evidence is now also emerging (21,23). Akinkugbe et al (16) recently reported that periodontitis was an independent risk factor for incident NAFLD, and

Yoneda et al (22) found that treatment of periodontitis led to reduction in serum transaminase levels.

Periodontitis could affect the liver by several potential mechanisms. First, in periodontitis, oral bacteria and locally produced proinflammatory cytokines can enter the blood stream and potentially induce liver damage (18,21,39)**.** Secondly, oral bacteria such as *Porphyromonas gingivalis* can cause alterations in the gut microbiota, with subsequent decreased expression of tight-junction proteins in the ileum and increased gut translocation and systemic inflammation (40,41). Bacterial translocation from the gut is a key mechanism in the progression of chronic liver disease (42).

The swallowed saliva of subjects with periodontitis can contain 10^9 bacteria/ml, and more than 10^{12} oral bacteria can enter the gut each day and potentially affect gut microbial composition (15,43). Interestingly, liver cirrhosis patients were found to have gut dysbiosis enriched with bacteria of oral origin (44). In another study, salivary dysbiosis was found to predict decompensation events among patients with liver cirrhosis (45). Periodontitis is a key driver of salivary dysbiosis (46,47). These data provide potential mechanistic explanation for the associations observed in our study.

The effect of periodontitis on the risk for liver disease was especially high in the subgroups of subjects with either NAFLD or heavy alcohol use at baseline. It is well known that only a small fraction of individuals with NAFLD or heavy alcohol use ever develop clinical liver disease, but the drivers of this risk are still poorly defined. Based on our findings, it could be speculated that the hepatic damage from periodontitis is particularly relevant in subjects with preexisting susceptibility (fatty liver). Supportive of this concept, in steatotic mice orally

infected with *Porphyromonas gingivalis*, Furusho et al (40) showed that *Porphyromonas gingivalis* becomes present in the steatotic liver and may promote hepatic inflammation.

Edentulous subjects differed markedly from the other groups by showing lower employment rates, lower education, less exercise, and a striking burden of metabolic risk. Under the premise that the reasons for tooth loss in this subgroup most likely included some prior experience of periodontitis (48), it is striking that this group throughout showed a lower risk of incident liver disease than the group with untreated, advanced periodontitis. This indirectly implies that treatment of periodontitis may reduce the risk for liver disease. Considerable uncertainty, however, remains to the risk estimates.

Treatment of periodontitis has been shown to improve oral dysbiosis and reduce systemic inflammation (49). However, whether or not treatment of periodontitis actually impacts the development of liver disease needs further study. Nonetheless, periodontitis is a preventable disease. Maintaining good oral health requires brushing teeth twice daily with fluoride toothpaste, regular interdental cleaning and having annual oral examinations by a dentist. Tobacco cessation is also important (50).

In conclusion, we found an epidemiological link between periodontitis and incident severe liver disease. This link was independent of multiple confounders. As a preventable disease, periodontitis might thus present a modifiable risk factor for chronic liver disease, but further study is needed.

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Figure legends

Figure 1. The relationship between number of ≥ 4 mm deep periodontal pockets and and risk for incident severe liver disease (hospitalization, liver cancer or liver death), by Cox regression analysis adjusted for age, sex, and number of teeth. Edentulous individuals are excluded. Dotted lines represent 95% confidence intervals to the relative risk (hazards ratio) estimate.

Table 1. Baseline demographics between the study groups.

	All subjects	Group A No periodontitis	Group B Mild to moderate periodontitis	Group C Advanced periodontitis	Group D Edentulous
Subjects	6165	1900	1610	1723	932
Age (years), mean (SD)	49.5 (13.2)	47.5 (13.3)	50.0 (13.2)***	51.4 (12.7)***	70.5 (11.4)***
Women, n (%)	3374 (54.7)	1198 (63.1)	845 (52.5)***	730 (42.4)***	601 (64.5)***
Marital status, n (%)					
Married/partnership	4288 (69.8)	1431 (75,6)	1161 (72.3)	1204 (70.1)***	492 (52.9)***
Single	1858 (30.2)	462 (24.4)	444 (27.7)	513 (29.9)	439 (47.1)
Education, n (%)					
Basic	2419 (39.4)	577 (30.5)	514 (32.0)	579 (33.7)***	749 (80.5)***
Secondary	1975 (32.1)	630 (33.3)	545 (34.0)	650 (37.9)	150 (16.1)
Higher	1749 (28.5)	685 (36.2)	545 (34.0)	487 (28.4)	32 (3.4)
Employment status, n (%)					
Part- or fulltime employed	3491 (56.8)	1270 (67.1)	1044 (65.0)**	1059 (61.7)***	118 (12.7)***
Retired	2003 (32.6)	379 (20.0)	384 (24.0)	468 (27.2)	772 (82.8)
Other	653 (10.6)	244 (12.9)	177 (11.0)	190 (11.1)	42 (4.5)
Diabetes, n (%)	561 (9.1)	127 (6.7)	125 (7.8)	135 (7.8)	174 (18.7)***
HOMA-IR, mean (SD)	2.4 (5.6)	2.1 (2.4)	2.3 (4.3)	2.7 (8.5)***	3.2 (3.8)***
Hypertension, n (%) ¥	3887 (63.3)	1061 (56.1)	943 (58.9)	1102 (64.1)***	781 (84.3)***
Metabolic syndrome, n (%) ¥¥	2752 (44.8)	719 (37.9)	641 (40.0)	796 (46.2)***	596 (64.3)***
Body mass index, mean (SD)	26.9 (4.6)	26.4 (4.7)	26.7 (4.4)*	27.1 (4.6)***	28.1 (5.0)***
GGT, U/L, mean (SD)	35.9 (44.9)	31.2 (40.1)	35.2 (43.4)***	40.6 (47.1)***	36.6 (49.9)***
Smoking, n (%)					
Current	1629 (26.5)	421 (22.2)	401 (25.0)	653 (38.0)***	154 (16.7)***
Former	1342 (21.9)	406 (21.5)	323 (20.1)	366 (21.3)	247 (26.6)
Never	3169 (51.6)	1065 (56.3)	880 (54.9)	698 (40.7)	526 (56.7)
Average alcohol consumption (g/week), mean (SD)	75.1 (144.6)	60.4 (107.2)	84.6 (158.4)***	105.6 (179.1)***	33.9 (96.8)***
Use of sugar-sweetened beverages, n (%)					
Seldom or never	3325 (55.5)	1032 (55.3)	866 (54.9)	946 (56.3)	481 (55.4)
2-5 times a week	1528 (25.5)	481 (25.8)	436 (27.7)	438 (26.1)	173 (19.9)
Daily	1136 (19.0)	352 (18.9)	274 (17.4)	295 (17.6)	215 (24.7)
Use of sweets daily, n (%)	229 (3.8)	53 (2.8)	39 (2.5)	50 (3.0)	87 (10.2)***
Exercise habits					
Twice or more weekly	3515 (58.8)	1107 (59.1)	963 (61.2)	963 (57.0)	482, (56.6)***
2-4 times in a month	1679 (28.0)	570 (30.4)	440 (27.9)	506 (30.0)	163 (19.2)
Rarely	793 (13.2)	196 (10.5)	172 (10.9)	219 (13.0)	206 (24.2)

¥ Blood pressure above 130/85 mmHg or antihypertensive medication
¥¥ According to the Joint Interim Statement Criteria [31], 3 of the following 5 metabolic syndrome components: hyperglycemia, elevated triglycerides, reduced HDL, increased waist circumference, and hypertension

Percentages are calculated within groups.
* P<0.05 compared to group A
**P≤0.01 compared to group A
***P≤0.001 compared to group A

Table 2. Periodontal health data between the groups.

	All subjects	Group A No periodontitis	Group B Mild to moderate periodontitis	Group C Advanced periodontitis
Subjects	6165	1900	1610	1723
No. of teeth, mean (SD)	22.9 (7.8)	22.4 (8.6)	22.6 (8.1)	23.8 (6.3)
No. of teeth with ≥4 mm pockets, mean (SD)			2.2 (1.1)	10.5 (5.3) **
Lower percentile			1	6
Median			2	9
Higher percentile			3	13
Dental examinations, n (%)				
Once a year	2170 (73.0)	777 (69.1)	702 (74.0)*	690 (77.0)**
Every two years	674 (22.7)	288 (25.6)	205 (21.6)	180 (20.1)
Less frequently	128 (4.3)	60 (5.3)	42 (4.4)	26 (2.9)
Tooth-brushing frequency, n (%)				
At least twice daily	3148 (62.3)	1201 (66.1)	971 (62.6)*	968 (58.2)**
Once daily	1551 (30.7)	537 (29.5)	487 (31.3)	521 (31.3)
Less often	352 (7.0)	80 (4.4)	94 (6.1)	175 (10.5)
Interdental cleaning, n (%)				
Daily	493 (9.9)	193 (10.7)	160 (10.4)	140 (8.6)**
Weekly	788 (15.8)	304 (16.8)	263 (17.1)	221 (13.5)
Less frequently	1081 (21.6)	426 (23.5)	326 (21.2)	328 (20.1)
Never	2636 (52.7)	887 (49.0)	788 (51.3)	946 (57.8)
Dental plaque, lower left lingual, n (%)				
No	2327 (48.0)	1064 (61.9)	745 (50.4) **	518 (31.3) **
Only at gumline	2162 (44.5)	586 (34.2)	661 (44.8)	915 (55.2)
Other places also	302 (6.2)	54 (3.1)	61 (4.1)	187 (11.3)
Bleeding on probing, upper right side				
No bleeding, n(%)	1935 (44.1)	960 (64.2)	641 (47.7) **	334 (21.6) **
Bleeding	2367 (53.9)	503 (33.7)	678 (50.4)	1186 (76.6)

Percentages are calculated within groups. Dental plaque and bleeding on probing -variables do not add up to 100%, some percentages within those groups were non-examinable.
* P<0.05 compared to group A
** P<0.001 compared to group A
# P<0.001

Table 3. Hazards ratios (HR) with 95% confidence intervals (CI) for incident severe liver events (hospitalization, liver cancer or liver death) according to periodontal health status by Cox regression analyses with various level of adjustment.

	Model 1	Model 2	Model 3	Model 4
	HR (95% CI)	HR (95% CI)	HR (95% CI)	HR (95% CI)
No periodontitis (Group A)	1 (reference)	1 (reference)	1 (reference)	1 (reference)
Mild to moderate periodontitis (Group B)	2.12 (0.98-4.58)	2.24 (1.00-5.02)*	2.35 (1.01-5.46)*	2.17 (0.98-4.84)
Advanced periodontitis (Group C)	3.69 (1.79-7.60)**	3.63 (1.67-7.86)**	3.72 (1.65-8.34)**	3.29 (1.53-7.05)**
Edentelous subjects (Group D)	1.48 (0.56-3.92)	1.48 (0.51-4.34)	n.c.	1.29 (0.45-3.68)

HR, Hazards Ratio

* P<0.05; ** P<0.005; n.c., non-calculable

Model 1: adjusted for age, sex, and number of teeth

Model 2: Model 1 + average alcohol use, waist circumference, diabetes, metabolic syndrome, HOMA-IR, GGT, and smoking status (current, former, never)

Model 3: Model 1 + average alcohol use, metabolic syndrome, teeth-brushing frequency, daily use of sweets, use of sweetened beverages, and exercise

Model 4: Model 1 + average alcohol use, metabolic syndrome, marital status, education level, and employment

Table 4. Hazards ratios (HR) with 95% confidence intervals (CI) for incident severe liver events (hospitalization, liver cancer or liver death) according to periodontal health status and in subgroups with baseline non-alcoholic fatty liver disease (NAFLD)[1], heavy alcohol use[2], or neither condition. Analyses are by Cox regression adjusted for age, sex, and number of teeth.

	Subjects	Liver events	HR	95%CI
No NAFLD, non-heavy drinker	3438	23		
No periodontitis (Group A)			1 (reference)	
Mild to moderate periodontitis (Group B)			1.85	0.58-5.89
Advanced periodontitis (Group C)			2.09	0.64-6.80
Edentulous subjects (Group D)			0.69	0.14-3.51
NAFLD[1]	1801	23		
No periodontal pockets (Group A)			1 (reference)	
Mild to moderate periodontitis (Group B)			3.23	0.62-16.8
Advanced periodontitis (Group C)			6.94	1.43-33.6*
Edentulous subjects (Group D)			1.34	0.24-7.68
Heavy alcohol use[2]	613	26		
No periodontitis (Group A)			1 (reference)	
Mild to moderate periodontitis (Group B)			4.74	0.58-38.6
Advanced periodontitis (Group C)			7.83	1.03-59.5*
Edentulous subjects (Group D)			4.11	0.19-91.3

[1] Fatty liver index >60 and average alcohol use <30 g/day for men and <20 g/day for women

[2] Average alcohol use ≥30 g/day for men and ≥20 g/day for women