Universitat Internacional de Catalunya
Facultat d'Odontologia

Tesis doctoral

Periodontitis and Blood Pressure:

Exploring the Association

Eva Mª Muñoz Aguilera

Periodontitis and Blood Pressure: Exploring the Association

Eva Mª Muñoz Aguilera

DOCTORAL THESIS
IN HEALTH SCIENCES

PERIODONTAL MEDICINE

Directors

Francesco D'Aiuto
Jose Nart

Universitat Internacional de Catalunya
Barcelona, July 2021

To my husband Hector,

"The world is richer in associations than meanings, and it is the part of wisdom to differentiate the two."

John Barth, novelist.

Acknowledgments

ACKNOWLEDGEMENTS

I would like to recognise that part of this work was carried out at the UCL Eastman Dental Hospital, that received approved funding from the NIHR (National Institute for Health Research).

I acknowledge that this PhD has been directed by Professor Francesco D'Aiuto and Dr Jose Nart, who I would like to express my sincere gratitude.

Prof. Francesco D'Aiuto has always encouraged me to pursue my dreams and guided every step of the way. I will be forever grateful to him for offering me an opportunity to learn from him and work under his wise umbrella.

Dr Jose Nart has provided unvaluable guidance and support for this thesis. He reached out when I needed his help and trusted this research was meaningful and worth efforts of broader collaborations.

I am really grateful to Dr Jeanie Suvan, who was always very enthusiastic of this research topic and encouraged me to go the extra mile.

Ana Di Lorio and Alex Stagg provided great support and guidance with the different search strategies of relevant literature for the systematic review.

My gratitude goes to Dr Jacopo Buti for his contribution with the meta-analysis of the systematic review and to Dr Queralt Miro Catalina, for her patience and support with my learning of health statistics and for her invaluable help with the mediation analysis.

I cannot forget my friends Marco and Yago, whom I have work alongside many hours and who offered wise pieces of advice and friendship. To all my other colleagues, Jason, Estela, Natalie, Fede, Prof. Needleman, Mr Patel, Mrs Hussain and Ms Darbar, and all other staff members at Eastman Dental Institute and Hospital.

To my friends Anjali, Husna, Eng Hong, and Divya, who have not stopped believing in me and cheering me up throughout this journey. They are truly friends for life.

To my lovely crew in Barcelona, Jose, Elia and Ali. For always being there for me and for providing such lovely memories of my time in Barcelona.

I am very blessed to have incredible friends in Spain, particularly Yoli, Laura, Anabel, Mari, Aurora, Lourdes, Davinia, Patricia, Armonía and Carmen were always available for me to reach out. They have all made tremendous efforts keeping in touch and allowing themselves to have time to meet anytime I visited Granada.

I could not have done this without the support, love and understanding of my humble and lovely family. I can thank them enough for their sacrifice and efforts to facilitating the opportunity given to pursue my dreams.

Last but not least, my most special thanks go to my beloved husband, Héctor, for his patience with the many hours of my absence, his unconditional love, support, and reassurance. For being so patient and caring and for always cheering me up when I was feeling low. He has been there in every step of the journey regardless of how many difficulties we faced.

Table of contents

TABLE OF CONTENTS

Abstract

ABSTRACT

Background: Recent evidence indicates a relationship between periodontitis and hypertension; however, the nature and magnitude of the association are yet to be determined. The aim of this PhD was to further our understanding of this relationship.

Material & Methods: To investigate the research hypothesis, three studies were conducted. A comprehensive systematic review (part 1) intended to ascertain the likelihood of hypertension in individuals with periodontitis when compared to those without periodontitis. Secondary objectives evaluated i: linear relationship between periodontitis (extent/severity) and hypertension [measures of blood pressure (BP)], ii: the mean BP values in periodontitis versus non-periodontitis and iii: the effect of periodontal therapy on systolic (SBP) and diastolic blood pressure (DBP). A cross-sectional analysis (part 2) of two well-established surveys of representative samples of USA and Korean populations was designed with to further assess, i: the degree of association between periodontitis and hypertension, ii: the potential role of systemic inflammation in mediating this association. A post-hoc nested case-control study (part 3) in a sample of systemically healthy participants with the objective to investigate i: the association between periodontitis and mean BP levels, ii: the relationship with systemic inflammatory makers and its mediation effect, iii: the level of possible undetected hypertension in the sample population.

Results: The systematic review included 81 studies (comprising observational and interventional designs). Moderate-severe periodontitis (OR = 1.22; 95% CI: 1.10–1.35, P=0.0001) and severe periodontitis (OR = 1.49; 95% CI: 1.09–2.05, P=0.01) were associated with hypertension. Diagnosis of periodontitis increased the likelihood of hypertension in prospective studies (OR = 1.68; 95% CI: 0.85–3.35, *P*=0.14). Further, participants with periodontitis exhibited higher mean SBP (WMD = 4.49 mmHg; 95% CI: 2.88–6.11, *P*=0.00001) and DBP (WMD = 2.03 mmHg; 95% CI: 1.25–2.81, *P*=0.00001) when compared to controls. Less than half (5 out of 12) of interventional studies confirmed a reduction in BP following the treatment of periodontitis, ranging from an average of 3-12.5 mm Hg of SBP and from 0-10 mm Hg of DBP.

The cross-sectional study results confirmed that participants with periodontitis had increased odds of hypertension (NHANES: OR = 1.3, 95% CI: 1.0–1.6, *P*=0.025;

KNHANES: OR = 1.2, 95% CI: 1.0–1.4, *P*=0.041) and actual SBP≥140 mmHg (NHANES: OR = 1.6, 95% CI: 1.1–2.3, *P*<0.001; KNHANES: OR = 1.3, 95% CI: 1.0–1.6, *P*<0.031) following adjustment of traditional confounders and confirmed in participants not taking antihypertensive medications. Diagnosis of periodontitis was directly associated with WBC in both surveys and with CRP levels in the NHANES. Mediation analyses confirmed that CRP acted as a mediator in the association between periodontitis and hypertension in both populations. WBC acted as a mediator in the KNHANES whilst in the NHANES, its effect was dependent of CRP inclusion in the model.

Lastly, from the nested case-control study cases (participants with severe periodontitis) presented with 3.36 mm Hg (95% CI, 0.91–5.82, *P*=0.007) higher mean SBP and 2.16 mm Hg (95% CI, 0.24–4.08, *P*=0.027) higher DBP than controls (without periodontitis). Diagnosis of periodontitis was associated with mean SBP (β=3.46±1.25, *P*=0.005) and greater odds of SBP≥140 mmHg (OR = 2.3, 95% CI, 1.15–4.60, *P*=0.018) independent of common cardiovascular risk factors. Similar findings were observed when continuous measures of periodontal status were modelled against SBP. Measures of systemic inflammation although elevated in periodontitis were not found to be mediators of the association between periodontitis and arterial BP values in this sample of participants without hypertension.

Conclusions: Diagnosis of periodontitis is consistently associated with increased odds of hypertension, higher SBP/DBP levels and systemic inflammatory biomarkers. Patient-centred approaches integrating both the medical and dental communities will be crucial in the prevention and management not only of periodontitis but also hypertension and its complications.

RESUMEN

Antecedentes: La evidencia actual sugiere una relación entre la periodontitis e hipertensión, sin embargo, la naturaleza y la magnitud de esta asociación están por determinar. Por lo tanto, esta tesis doctoral tiene el objetivo de discernir su relación.

Material y métodos: Para estudiar la hipótesis de la investigación, tres metodologías fueron llevadas a cabo. La revisión sistemática de la literatura (1ª parte), cuyo objetivo primario fue investigar si pacientes con periodontitis presentaban un mayor riesgo de hipertensión en comparación con aquellos sin periodontitis. Los objetivos secundarios evaluaron i: la relación lineal de la periodontitis (extensión/severidad) con la hipertensión [medidas de presión arterial (PA)], ii: los valores medios de la PA en periodontitis versus no periodontitis, iii: el efecto del tratamiento periodontal en la PA sistólica (PAS) y diastólica (PAD). El estudio transversal (2ª parte) de dos bases de datos representativas de las poblaciones norteamericana y coreana tuvo como objetivos i: corroborar los resultados de la revisión sistemática en cuanto al riesgo de hipertensión en pacientes con periodontitis y ii: analizar la asociación con la inflamación sistémica y su posible efecto mediador. El estudio de casos y controles (3ª parte) se diseñó con el objetivo de investigar i: la asociación entre periodontitis y los valores medios de PAS/PAD, ii: la relación con los marcadores de inflamación sistémica y su posible efecto mediador, iii: ll porcentaje de hipertensión no diagnosticada en la población de estudio.

Resultados: La revisión sistemática incluyó 81 estudios (observacionales y de intervención). La periodontitis moderada a severa (OR = 1.22; 95% CI: 1.10–1.35, *P*=0.0001) y la periodontitis severa (OR = 1.49; 95% CI: 1.09–2.05, *P*=0.01) se asociaron a hipertensión. Además, los estudios longitudinales de cohortes confirmaron los resultados; la periodontitis incrementó el riesgo de hipertensión (OR = 1.68; 95% CI: 0.85–3.35, *P*=0.14). Los valores de PAS (WMD = 4.49 mmHg; 95% CI: 2.88–6.11, *P*=0.00001) y PAD (WMD = 2.03 mmHg; 95% CI: 1.25–2.81, *P*=0.00001) se encontraron más elevados en pacientes periodontales en comparación con aquellos sin la enfermedad. Por último, solamente 5 de 12 estudios de intervención confirmaron una reducción en PAS=3-12.5 mmHg y de PAD=0-10 mmHg tras el tratamiento periodontal. Los resultados del estudio transversal mostraron que participantes con periodontitis tenían un riesgo de hipertensión aumentado (NHANES: OR = 1.3, 95% CI: 1.0–1.6,

P=0.025; KNHANES: OR = 1.2, 95% CI: 1.0–1.4, *P*=0.041) y además, de PAS≥140 mmHg (NHANES: OR = 1.6, 95% CI: 1.1–2.3, *P*<0.001; KNHANES: OR = 1.3, 95% CI: 1.0–1.6, *P*<0.031) después de ajustar las variables de confusión. Los resultados se confirmaron en aquellas personas que no tomaban medicación antihipertensiva. La periodontitis estaba directamente asociada con el recuento de glóbulos blancos (RGB) en las dos bases de datos y con la proteína C-reactiva (PCR) en NHANES. Los análisis de mediación confirmaron que la PCR medió la relación entre periodontitis e hipertensión en las dos poblaciones. El RGB actuó como mediador en KNHANES mientras que en NHANES, el efecto de mediación dependió de la inclusión de la PCR en el modelo.

Los resultados del estudio de casos y controles mostraron que los individuos con periodontitis severa (casos) presentaron una PAS [3.36 mm Hg (95% CI, 0.91–5.82, *P*=0.007)] y PAD [2.16 mmHg (95% CI, 0.24–4.08, *P*=0.027)] más elevada que los controles (individuos sin periodontitis). La periodontitis estaba asociada con la PAS [variable continua (β=3.46±1.25, *P*=0.005)] y un riesgo más elevado de tener PAS≥140 mmHg (OR = 2.3, 95% CI, 1.15–4.60, *P*=0.018), independientemente de variables cardiovasculares comunes. Se obtuvieron resultados similares cuando el modelo se realizó con variables periodontales continuas y PAS. Las medidas de inflamación sistémica (PCR y RGB) se encontraron elevadas en pacientes con periodontitis, pero no actuaron como mediadoras de la asociación entre periodontitis y los valores de PA.

Conclusiones: La periodontitis está asociada con un mayor riesgo de padecer valores de PA elevada e hipertensión, así como también de marcadores de inflamación sistémica, los cuales podrían actuar como mediadores de la asociación. La integración de estrategias de salud por parte de las comunidades médica y dental con la atención centrada en el paciente son clave tanto en la prevención como en el control de la periodontitis e hipertensión con el objetivo de disminuir el riesgo cardiovascular.

L st of p bl cat ons

LIST OF ORIGINAL PUBLICATIONS

This PhD is based on the three following manuscripts:

1st Publication

Reference: Muñoz Aguilera E, Suvan J, Buti J, Czesnikiewicz-Guzik M, Barbosa Ribeiro A, Orlandi M, et al. Periodontitis is associated with hypertension: a systematic review and meta-analysis. Cardiovascular research. 2020;116(1):28-39.

Title: Periodontitis is associated with hypertension: a systematic review and meta-analysis.

Authors: Muñoz Aguilera, E., Suvan, J., Buti, J., Czesnikiewicz-Guzik, M., Barbosa Ribeiro, A., Orlandi, M., Guzik, T.J., Hingorani, A.D., Nart, J. and D'Aiuto, F.

Journal: Cardiovascular Research (IF:8.168) https://doi.org/10.1093/cvr/cvz201

2nd Publication

Reference: Muñoz Aguilera E, Leira Y, Miró Catalina Q, Orlandi M, Czesnikiewicz-Guzik M, Guzik TJ, et al. Is systemic inflammation a missing link between periodontitis and hypertension? Results from two large population-based surveys. Journal of Internal Medicine. 2021;289(4):532-46.

Title: Is Systemic Inflammation the Missing Link Between Periodontitis and Hypertension? Results from Two Large Population-based Surveys.

Authors: Muñoz Aguilera, E., Leira, Y., Miró Catalina, Q., Orlandi, M., Czesnikiewicz-Guzik, M., Guzik, T.J., Hingorani, A.D., Nart, J. and D'Aiuto, F.

Journal: Journal of Internal Medicine (IF: 6.871) https://doi.org/10.1111/joim.13180

3rd Publication

Reference: Muñoz Aguilera E, Suvan J, Orlandi M, Miró Catalina Q, Nart J, D'Aiuto F. Association between periodontitis and blood pressure highlighted in systemically healthy individuals: Results from a nested case-control study. Hypertension. 2021;77(5):1765-74.

Title: Association between periodontitis and blood pressure highlighted in systemically healthy individuals. Results from a nested case-control study

Authors: Muñoz Aguilera, E., Suvan, J., Orlandi, M., Miró Catalina Q Nart, J., and D'Aiuto, F.

Journal: Hypertension (IF.7.713)

https://doi.org/10.1161/HYPERTENSIONAHA.120.16790

LIST OF SUPLEMENTARY ORIGINAL PUBLICATIONS

In line with the PhD research topic, the following manuscripts add further insight and broaden this doctoral thesis. These were carried out in collaboration with researchers in Portugal, Scotland, Poland and Italy during the PhD timeline. As a result, a wider and stronger team is developing aimed at enhancing the quality of its research and dissemination of its outcomes.

1st Publication (IF:22.637)

Reference: Czesnikiewicz-Guzik M, Osmenda G, Siedlinski M, Nosalski R, Pelka P, Nowakowski D, et al. Causal association between periodontitis and hypertension: evidence from Mendelian randomization and a randomized controlled trial of non-surgical periodontal therapy. European heart journal. 2019;40(42):3459-70. https://doi.org/10.1093/eurheartj/ehz646

2nd Publication (IF: 5.688)

Reference: Machado V, Aguilera EM, Botelho J, Hussain SB, Leira Y, Proença L, et al. Association between periodontitis and high blood pressure: results from the study of periodontal health in Almada-Seixal (SoPHiAS). Journal of clinical medicine. 2020;9(5):1585. https://doi.org/10.3390/jcm9051585

3rd Publication (IF: 4.171)

Reference: Pietropaoli D, Monaco A, D'Aiuto F, Aguilera EM, Ortu E, Giannoni M, et al. Active gingival inflammation is linked to hypertension. Journal of hypertension. 2020;38(10):2018-27. https://doi.org/10.1097/HJH.0000000000002514

4th Publication (IF: 1.26)

Reference: Del Pinto R, Pietropaoli D, Munoz-Aguilera E, D'Aiuto F, Czesnikiewicz-Guzik M, Monaco A, et al. Periodontitis and hypertension: is the association causal? High Blood Pressure & Cardiovascular Prevention. 2020;27:281-9. https://doi.org/10.1007/s40292-020-00392-z

5th Publication (IF: 5.893)

Reference: Sharma S, Sridhar S, McIntosh A, Messow C-M, Aguilera EM, Del Pinto R, et al. Periodontal therapy and treatment of hypertension-alternative to the pharmacological approach. A systematic review and meta-analysis. Pharmacological Research. 2021;166:105511. https://doi.org/10.1016/j.phrs.2021.105511

Introduction

1. INTRODUCTION

1.1. AIM

This PhD aimed to investigate the nature of relationship between periodontitis and hypertension and explore further the role of systemic inflammation in mediating the association.

1.2. BACKGROUND

The background describes the research topics, with an interpretation of the available literature in the context of the possible links between periodontitis and hypertensin and discusses the rationale for this thesis.

1.2.1. HYPERTENSION

DEFINITION AND PREVALENCE

Hypertension is a non-communicable chronic disorder wherein blood pressure in the arteries is persistently elevated. Two measures can be conveyed, namely systolic blood pressure (SBP) or the highest blood pressure during contraction of the ventricles and diastolic blood pressure (DBP), relating to the lowest blood pressure noted preceding the next contraction (1, 2). Hypertension was defined by the Sixth Report of the Joint National Committee on Prevention, Detection, Evaluation, and Treatment (JNC VI) as the condition associated with values of SBP $\geq$ 140 mm Hg and/or DBP $\geq$ 90 mm Hg in individuals who are not taking antihypertensive medications (3). This definition and the choice of these thresholds of BP was mostly based on the evidence derived from interventional RCTs confirming the benefits in reducing blood pressure in terms of cardiovascular outcomes and complications (4).

Essential or primary hypertension is the commonest form of the disease, whilst other types are described such as secondary and resistant hypertension, elderly or pregnancy-

related forms, hypertension within metabolic syndrome, malignant hypertension and white coat hypertension, among others (4).

Measurements of blood pressure are highly prone to error, thus, to confirm hypertension diagnosis, more than a single set of BP readings (1-2 min apart) at different office visits (depending on severity) and/or home blood pressure and/or ambulatory blood pressure monitoring are required (5). It is now recommended that standardized oscillometer devices are used for measuring blood pressure due to their improved accuracy compared with conventional auscultatory methods (6). Current case definitions of hypertension using different SBP and DBP thresholds vary across continents but they could be described when examining the recent European (7) and American guidelines (8).

Guideline Differences	American College of Cardiology/American Heart Association (ACC/AHA)		European Society of Cardiology/European Society of Hypertension (ESC/ESH)	
Level of blood pressure (BP) defining hypertension	Systolic (mm Hg) and/or	Diastolic (mm Hg)	Systolic (mm Hg) and/or	Diastolic (mm Hg)
Office/Clinic BP	≥ 130	≥ 80	≥ 140	≥ 90
Daytime mean	≥ 130	≥ 80	≥ 135	≥ 85
Nighttime mean	≥ 110	≥ 65	≥ 120	≥ 70
24-hour mean	≥ 125	≥ 75	≥ 130	≥ 80
Home BP mean	≥ 130	≥ 80	≥ 135	≥ 85
BP targets for treatment	< 130/80 mm Hg		Systolic targets < 140 mm Hg and close to 130 mm Hg	
Initial Combination Therapy	Initial single-pill combination therapy in patients > 20/10 mm Hg above BP goal		Initial single-pill combination therapy in patients ≥ 140/90 mm Hg	
Hypertensive requiring intervention	> 130/80 mm Hg		≥ 140/90 mm Hg	

Guideline Similarities	ACC/AHA	ESC/ESH
Importance of home BP monitoring	• Take BP at home, twice in the morning and twice in the evening, in the week before clinic • Bring the BP machine in annually for validation	
Therapy	• Restrict beta blockers to patients with comorbidities or other indications • Initial single pill combination as initial therapy	
Follow-up	• Detect poor adherence and focus on improvement • BP telemonitoring and digital health solutions recommended	

Table 1. Comparison of categories of blood pressure according to current clinical guidelines. Reproduced with permission from Bakris et al. 2019 (9)

Hypertension is the most widespread of all cardiovascular disorders with 30–45% of the worldwide adult population affected by some forms of the disease and it represents approximately 1.13 billion people across the globe according to reports in the year 2015 (10). Its prevalence increases progressively with ageing, involving > 60% of individuals over 60 years old, independent of income status (11). Male individuals as well as ethnic minorities such as blacks are particularly susceptible to hypertension with a 10-15% higher prevalence than other ethnic groups (11, 12).

AETIOLOGY AND PATHOGENESIS OF HYPERTENSION

Hypertension is a highly heterogeneous condition with a multifactorial aetiology (13). Despite the wealth of evidence collected over the last 30 years, however the pathophysiology of this condition remains unclear. It is often labelled as essential hypertension, whereby no clear single identifiable cause is identified, whilst renal or adrenal disorders are the most common underlying causes for a minority of patients (between 2% and 5%) (14).

Individuals with elevated blood pressure often present with a positive family history of hypertension, with most studies attributing an estimated 35% and 50% of heritability (15). Genetic analysis of more than 1 million persons of European descent identified 535 new loci associated with hypertension traits (16). In addition, several risk factors such as being overweight and sedentary lifestyle, diabetes, diet imbalances with low calcium and potassium but rich in salt and alcohol consumption, smoking, aging and stress, are recognised to increase the odds of hypertension (17).

The development of essential hypertension has been associated with cardiac, vascular and/or renal systems derangements (Table 2) (14). Overactivation of the regulatory systems involving salt-intake and cardiovascular function (i.e. the renin-angiotensin-aldosterone and the sympathetic nervous systems) have been traditionally linked to hypertension development. Nevertheless, therapies targeting those imbalances have failed to control hypertension in up to 44-66% of the cases, suggesting other mechanisms causing hypertension are involved (18).

Physiological mechanisms involved in regulating blood pressure
1. Cardiac output
2. Peripheral resistance
3. Renin-angiotensin-aldosterone system
4. Autonomic nervous system
5. Other factors:
Bradykinin
Endothelin
EDRF (endothelial derived relaxing factor) or nitic oxide
ANP (atrial natriuretic peptide)
Ouabain

Table 1. Mechanisms altered in hypertension. With permission from Beevers et al. 2001 (14).

Current evidence indicates that systemic inflammation is linked to hypertension and that oxidative stress and endothelial dysfunction are among the main mechanisms involved (Figure 1) (19). Although inflammation provides a crucial role in the defense mechanism against any harmful stimuli, newer evidence confirm that persistent inflammation can have detrimental effects especially on the vasculature. In particular immunological dysfunctions have been implicated in the development of hypertension (20). Several immune cells have been involved in the pathogenesis of hypertension and hypertensive end-organ damage such as T and B lymphocytes cells, dendritic cells, and macrophages (21).

The first experimental (rat animal) model demonstrated that inhibition of infiltration and proliferation of immune cells, particularly T lymphocytes contributed to renal changes and prevention of BP elevation (22). Early clinical studies of vascular inflammation suggested Th17 cells subtype contributed to vascular dysfunction and hypertension development (23). A clinical study showed a relationship between vascular wall stiffness in untreated hypertensive patients with increased levels of acute phase proteins such as C-reactive protein (CRP) (24). Moreover, inflammatory processes can result in an overexpression of endothelial cell activation markers including vascular cell adhesion molecule-1 (VCAM-1) and intercellular adhesion molecule-1 (ICAM-1) which in turn have been implicated in hypertension development and predict future CV events (25).

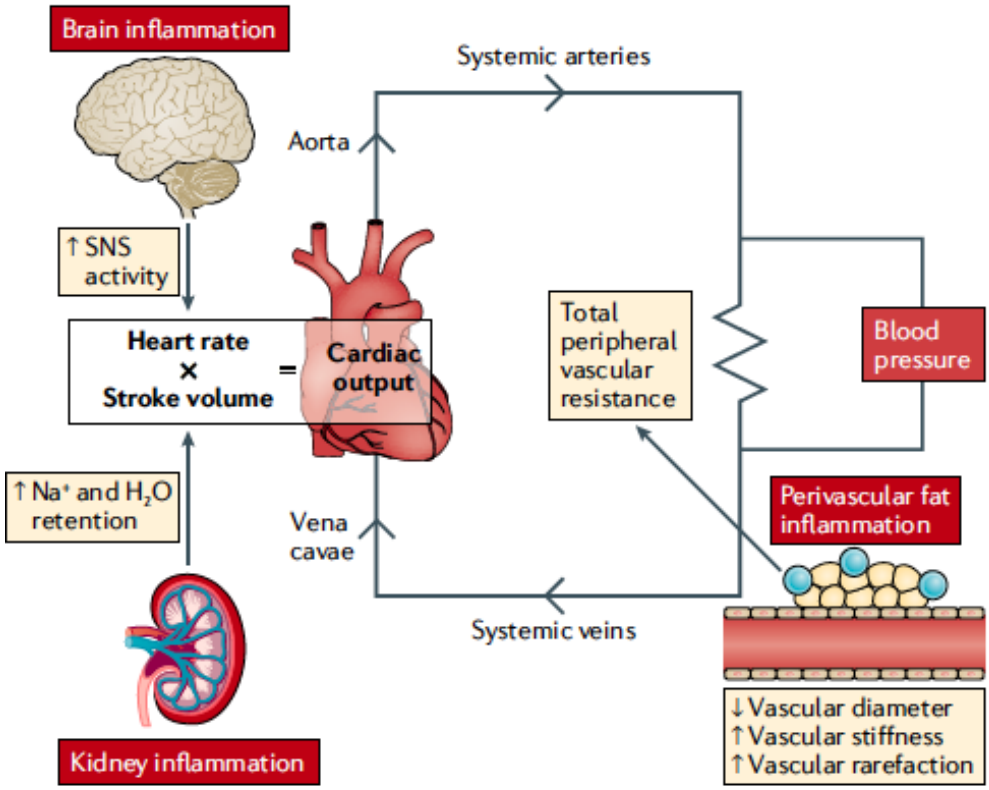


Figure 1. Inflammation altering the main tissues regulating blood pressure (with permission from Drummond et al. 2019) (26).

HYPERTENSION RISK FOR CARDIOVASCULAR DISEASES

Hypertension is identified as the major risk factor for cardiovascular morbidity, mortality and disability worldwide (27). A strong linear positive correlation has been reported between elevated office blood pressure and risk of future cardiovascular incidents (heart failure, stroke, myocardial infarction, sudden death and peripheral artery disease), and renal diseases, with observational studies showing a progressive increase in cardiovascular risk over 115 mm Hg of SBP (4, 28). In 2015 diagnosis of hypertension accounted for almost 10 million deaths and over 200 million disability-adjusted life years worldwide (29).

MANAGEMENT OF HYPERTENSION.

The ultimate goal of anti-hypertensive therapies is to reduce the risk of cardiovascular complications. Lowering arterial blood pressure more than 10 mm Hg in systolic (SBP)

and more than 5 mmHg in diastolic (DBP) produces a significant reduction of cardiovascular events (30). Indeed, experimental evidence from randomised controlled trials confirms that SBP/DBP reductions below 140/90 mm Hg achieved a 35-40% reduction in incident stroke, 15-20% reduction in myocardial infarction and 64% reduction in heart failure (3, 31).

Appropriate lifestyle changes can help not only in preventing the occurrence of hypertension in normotensive individuals, but they can reduce mean blood pressure values, the number of medications and/or doses in those individuals already taking antihypertensive medications. These measures recommended by the 2013 ESH/ESC guidelines comprise: attain a healthier lifestyle with reduction of salt and alcohol intake, favouring low-fat and carbohydrates, weight loss and smoking cessation, while increasing fruit and vegetables consumption, as well as regular physical exercise (4). Nevertheless, poor long-term adherence to these actions is perhaps the greatest limitation of these non-pharmacological interventions.

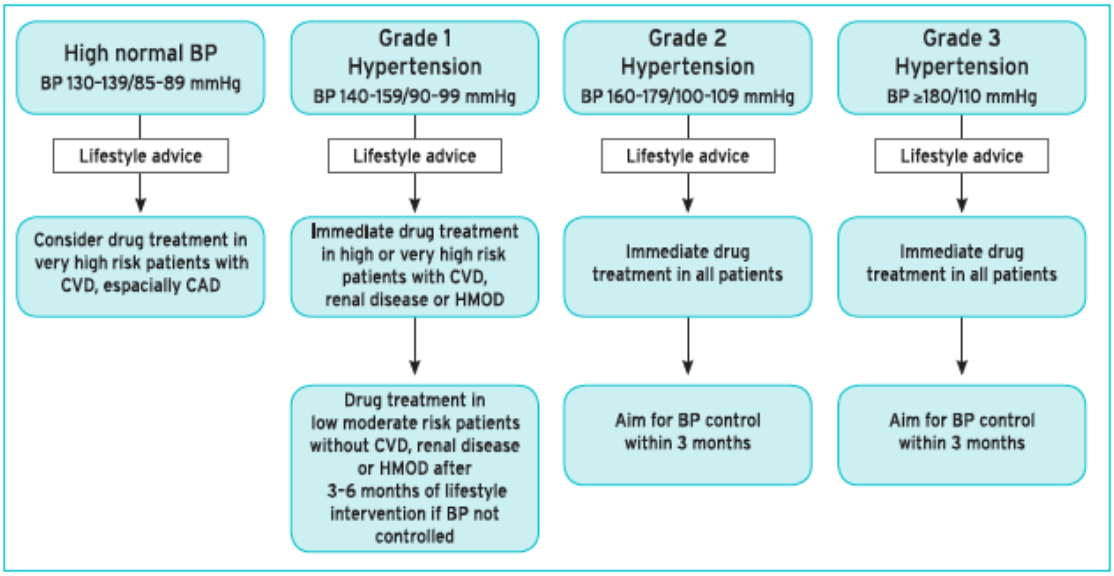


Figure 2. Pharmacological and non-pharmacological interventions according to disease severity. BP, blood pressure; CAD, coronary artery disease; CVD, cardiovascular disease; HMOD, hypertension-mediated organ damage. Reproduced with permission from Williams *et al.* 2018 (7).

The use of antihypertensive drugs has been recommended in patients with SBP/DBP of ≥140/90 mm Hg when non-pharmacological interventions have proven unsuccessful.

Further those individuals with a lower threshold of BP (130-139/85-89 mm Hg) but with added risk factors and cardiovascular risk (i.e. diabetes, previous cardiovascular or kidney diseases) have been included in the groups of patients who could benefit from pharmacologic blood-pressure lowering (4). Current drug treatments are directed towards limiting the influence of the renin-angiotensin-aldosterone and sympathetic nervous systems on blood pressure. The main groups commonly used either as a monotherapy or combination thereof are i: diuretics, ii: adrenoceptor antagonists, iii: angiotensin receptor blockers, iv: angiotensin converting enzyme (ACE) inhibitors, v: calcium channel antagonists, and vi: β-blockers. Despite the relatively wide range of pharmacologic treatments, only one third of those receiving blood pressure medications manage to achieve the therapeutic targets (11).

1.2.2. PERIODONTITIS

DEFINITION AND PREVALENCE

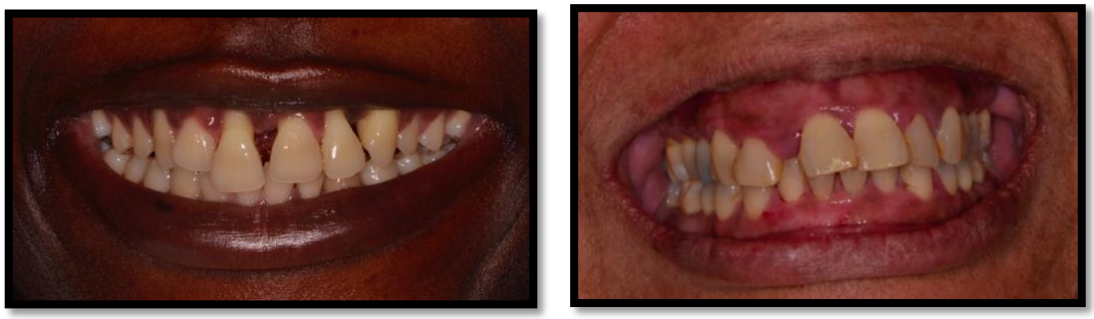
Periodontitis is a chronic multifactorial inflammatory disease initiated by a dysbiosis of the oral microbiota and associated with a dysfunctional inflammatory process leading to progressive destruction of the teeth supporting structures (32, 33). It is in most cases a preventable and treatable condition, although if left to progress it will eventually lead to tooth loss. Case definitions of periodontitis include mild (Stage I), moderate (Stage II) or severe (Stage III/IV) forms of periodontitis depending on different severity and complexity parameters defined in the latest classification (34) (Table 3).

Periodontitis is a major public health concern that involves almost 750 million people worldwide, with its severe form (7.4-11.2% of the population) named the 6th most prevalent disease and milder forms affecting over 50% of adults (35-37). The prevalence of periodontitis increases with age and male gender is often more affected than female, mostly related to societal or lifestyle factors rather than inherent differences in susceptibility between genders (38, 39). Current evidence also implies that severe periodontitis does not have an even representation for persons with certain ethnicities and socioeconomic backgrounds (40).

Periodontitis stage		Stage I	Stage II	Stage III	Stage IV
Severity	Interdental CAL at site of greatest loss	1 to 2 mm	3 to 4 mm	≥5 mm	≥5 mm
	Radiographic bone loss	Coronal third (<15%)	Coronal third (15% to 33%)	Extending to middle or apical third of the root	Extending to middle or apical third of the root
	Tooth loss	No tooth loss due to periodontitis		Tooth loss due to periodontitis of ≤4 teeth	Tooth loss due to periodontitis of ≥5 teeth
Complexity	Local	Maximum probing depth ≤4 mm Mostly horizontal bone loss	Maximum probing depth ≤5 mm Mostly horizontal bone loss	In addition to stage II complexity: Probing depth ≥6 mm Vertical bone loss ≥3 mm Furcation involvement Class II or III Moderate ridge defect	In addition to stage III complexity: Need for complex rehabilitation due to: Masticatory dysfunction Secondary occlusal trauma (tooth mobility degree ≥2) Severe ridge defect Bite collapse, drifting, flaring Less than 20 remaining teeth (10 opposing pairs)
Extent and distribution	Add to stage as descriptor	For each stage, describe extent as localized (<30% of teeth involved), generalized, or molar/incisor pattern			

Table 3. Periodontitis staging (Reproduced with permission from Tonetti *et al.* 2018)

Periodontitis and its sequelae are well known to worsen oral health and aesthetics, as it is associated with masticatory dysfunction, altered speech, with an impact on self-esteem and poorer quality of life (41-43) (See Picture 1). Further, periodontitis brings about social inequality and disability and it considerably increases healthcare costs (44, 45).



Picture 1. Clinical extraoral photographs (smile) of two women with generalised periodontitis (Stage IV, grade C).

AETIOLOGY AND PATHOGENESIS OF PERIODONTITIS

The understanding of periodontal diseases aetiology and pathogeneiss has increased continuously over the last 70 years. Löe and co-workers, on the well-known study on experimental gingivitis in man confirmed that accumulation of the dental biofilm consistently led to gingivitis formation, a reversible inflammation of the gingival tissues affecting the soft tissue compartment of the periodontium (46). Nevertheless, only in certain susceptible individuals this gingival inflammation would develop into periodontitis, concomitant with irreversible attachment loss and bone destruction (47). Certain microorganisms act as causative agents and have been strongly linked to periodontitis, but these pathogens can also be present in individuals with a healthy periodontium (48, 49). Bacteria are necessary for the disease development, but the mere presence of certain pathogens does not guarantee periodontitis progression, as it is the host inmuno-inflamatory response the determinant factor behind the progression from early gingival inflammatin to periodontal destruction (50).

Our current understanding of the pathogensis of periodontitis is based on the presence of specific bacteria or "keystone-pathogens", regardless of counts but that can turn a nonthreatening microbiota into a pathogenic one, altering the diversity, richness and proportions of its species (51, 52). Different members of this rich polymicrobial community seem to have distinctive roles, but contribute synergistically to trigger inflammatory porcesses and tissue destruction in periodontally-susceptible hosts (53). Moreover, a homeosthatic equilibrium that exists in health conditions between the host and different microorganisms can be disrupted by several factors that effect increasing susceptibility to periodontitis and its progression. Among them, insuline resistance, host immunodeficiencies, obesity, aging, social factors such as smoking, unhealthy diet, and increased stress, and other envirormental epigenetic adaptations (33, 54).

Bacterial aggressions triger an extremely organised chornological immune-inflammatory response aimed at protection from infections and to avoid damage to the host (55). The innate immunity represents the first barrier against bacterial challenges or other agreesions; it is indeed an effective system to restrain most pathogens from an

established infection (56). Bacteria and their products will trigger cytokines release by epithelial cells and activation of the complement (C3a-C5a), resulting in increased vascular permeability and leakage of neutrophils from the gingival vassels (57). The production of this inflammatory exudate is essential for non-specific defense mechanisms (55). Subsequently, monocytes/macrophages will access the site for detection and phagocytosis of bacteria, debris and apoptotic neutrophils to avoid prolonging inflammation (58). The inflammatory response to microorganisms is protective, nonetheless the acute inflammatory lesion turns into a chronic inflammation when mechanisms fail to remove neutrophils products and extracelular traps and other apoptotic inflammatory cells (59). It has been hypothetised that failure to activate inflammation resolution programs is associated to an exacerbated inflammatory response, which perpetuates damage in the host and caracterised the prolonged inflammation observed in periodontitis (59).

Innate recognition promotes an adaptive immune response that involves cellular and humoral mechanisms to a variety of triggers, it is a slower but more effective defensive process. The period of transition from the innate to the acquired immune response relates to the established periodontal lesion histologically, where macrophages, plasma cells, and lymphocytes T and B become the dominant cell infiltrate in parallel with the shift from moderate to severe gingival inflammation (60).

Essential in the orchestration of an effective and self-containing immune response is the ability of the host to recognize possible external aggressions. Bacterial pathogens or pathogen-associated molecular patterns (PAMPs) are indeed recognized by Toll-like receptors (TLR) in denditric cells, PMNs, macrophages, osteoclast precursos and osteoblasts. Binding of TLR induces the production of interleukin-1 (IL1), interleukin-6 (IL6), tumour necrosis factor alpha (TNFα) and prostaglandin E2 (PGE2) (61). These pro-inflammatory cytokines induce fibroblasts and other local cells to produce matrix metalloproteinases (MMPs) triggering extracellular matrix degradation (62). This torrent of inflammatory response also impact on the interplay between osteoblasts-osteoclasts balance with an increased osteoclastic activity fostering alveolar bone resorption (63).

Different TLR are associated with specific purposes in innate response and also in the origination of different T-cell subsets (64). Specificity of activation of subclasses of T cells could lead to either a cell-mediated immunity, protective antibody response (Th1) and contained lesion, or to a non-protective antibody response, with polyclonal B-cell activation (Th2) and progressive periodontal lesion (65). An important role of T helper 17 (Th17) and IL-17 cytokines has been documented, with induction of expression of receptor activator of nuclear factor-κB ligand (RANKL) in osteoblasts, and osteoclast activation and bone resorption upon interaction with its receptor activator nuclear factor (RANK) in pre-osteoclats (66, 67).

Additionally, leucocytosis and acute-phase proteins are produced when these local pro-inflamatory mediators enter the blood stream. With continued exposure, soluble antigens react with circulating specific antibody to form immune complexes that further amplify inflammation at local and other extraoral sites (57).

ORAL SYSTEMIC LINK

In the first part of the twentieth century, the theory of oral sepsis was described, which connected oral infections with "almost every system of the body" (68). Ever since, numerous studies have documented the impact of periodontal infection/inflammation on general health outcomes, with more than 57 systemic diseases and conditions hypothetised to be associated with periodontal diseases (69). Stronger epidemiological and clinical evidence is available for diabetes mellitus, cardiovascular diseases (CVD), and complication during pregnancy, independent of common risk factors such as smoking and obesity (70-73). The most commonly described plausible pathophysiological mechanisms underlying these associations involve a direct pathway with bacteraemia and endotoxaemia from the ulcerated epithelial lining of the periodontal pocket and an indirect pathway with inflammatory mediators from periodontal tissues entering the bloodstream (74). Porphyromonas gingivalis and Actynomyces actinomycetemcomitans are recognised periodontal pathogens that have been detected viable in atherosclerotic lesions, whereby could also act as pro-atherogenic stimuli (75). Increasing circulatory levels of pro-inflammatory cytokines (IL-1 beta, IL-2, IL-6 and IL-8), fibrinogen, serum

amyloid, leukocytosis and acute phase CRP induction from the liver have been linked to periodontitis (76-78). Direct and indirect pathways have been implicated in linking poro oral health and systemic health complications. Low-grade chronic systemic inflammation could result in endothelial dysfunction and atherosclerosis. On the other hand a dysbiosis of the gut microbiota, immune suppresion and bone marrow modifications have also been implicated as novel patways linking periodontitis to other systemic comorbidities (74). Lastly, translocation of periodontal bacteria via aspiration or through the digestive route has also been described as a direct pathway linking to respiratory complications like pneumonia but also in gut microbiome dysbiosis (79). Further research is warranted to indentify the exact mechanisms linking periodontitis to systemic health outcomes.

MANAGEMENT OF PERIODONTITIS

Treatment of periodontitis is aimed at arresting the progression of periodontitis, prevention of tooth loss and long-term preservation of the dentition in absence of signs and symptoms of the disease. Comprehensive mechanical removal of the dental biofilm in combination with self-performed dental hygiene either with non-surgical or surgical therapies, and regular supportive maintenance care are the cornerstones of successful treatment of periodontitis and attainment of periodontal health (80, 81). Accepted clinical endpoints include shallow gingival probing pocket depths (i.e. gingival pocket closure $\leq$ 4 mm) and stabilization of attachment levels, concomitant with low gingival bleeding and dental plaque scores (82, 83). These clinical endpoints correlate with reduction of inflammatory cells and gingival crevicular fluid flow, with improved healing of the connective tissue matrix accompanied by increased number of fibroblasts and collagen fibers along with a swift in the subgingival environment, involving quantitative and compositional declines of the periodontal pathogens (84, 85).

An ever-increasing number of studies suggest that successful treatment of periodontitis could achieve not only oral and periodontal health but also contribute to improve systemic health outcomes including prevention and control of metabolic conditions, cardiovascular diseases, and pregnancy complications (86-88). A reduction of systemic markers of inflammation and cardiometabolic risk factors, as well as

improvement of endothelial dysfunction and vasodilatation have been consistently described in patients with periodontitis (89-92). Moreover, studies have reported a reduction of future cardiovascular in refractory hypertensive patients (93, 94). An early randomised controlled clinical trial proposed that effective treatment of severe generalised periodontitis in otherwise healthy individuals was associated with a decrease in blood pressure after 2 months of therapy (95).

It has been documented that untreated periodontitis increases costs for non-oral medical conditions and patient hospitalization rates (96). On the other hand, favourable outcomes have been achieved for improving oral health related quality of life (OHRQoL) upon successful non-surgical treatment of periodontitis, whereas ineffective treatment correlated to poorer OHRQoL outcomes (43, 97). A recent systematic review claimed that OHRQoL improvements following non-surgical management of periodontitis can be achieved shortly after provision of the treatment (approximately at 1-2 weeks) maintained at least after 3 months (98).

1.2.3. **BIOLOGICAL PLAUSIBILITY**

Over the last decade, emerging evidences pointed at a strong relationship between periodontitis and hypertension (99, 100). These two common diseases share several risk factors such as smoking, diabetes, stress, obesity, increased age, as well as socioeconomic aspects (101). Subsequently, questions arise whether the relationship between periodontitis and hypertension might be blurred by common risk factors. Current evidence supports the notion that periodontitis is linked to cardiovascular diseases independent of traditional risk factors (70, 102).

Several pathways have been described supporting the association between periodontitis and hypertension although the exact mechanisms are yet to be fully understood (Figure 3) (103, 104). Firstly periodontitis could add to the derangement of the various systems involved in the pathogenesis of hypertension by contributing to the activation of host inflammation and rise of vasoactive endothelial substances (105). The increased level of serum CRP observed in patients with periodontitis may be a marker of

systemic inflammation which could in turn impact on hypertension (77, 106). Periodontitis is also characterised by a large oxidative stress burden both at the gingival and systemic level with a putative effect on endothelial function through vasculature changes (resistance and volume) leading to arterial stiffness and BP elevation (107, 108).

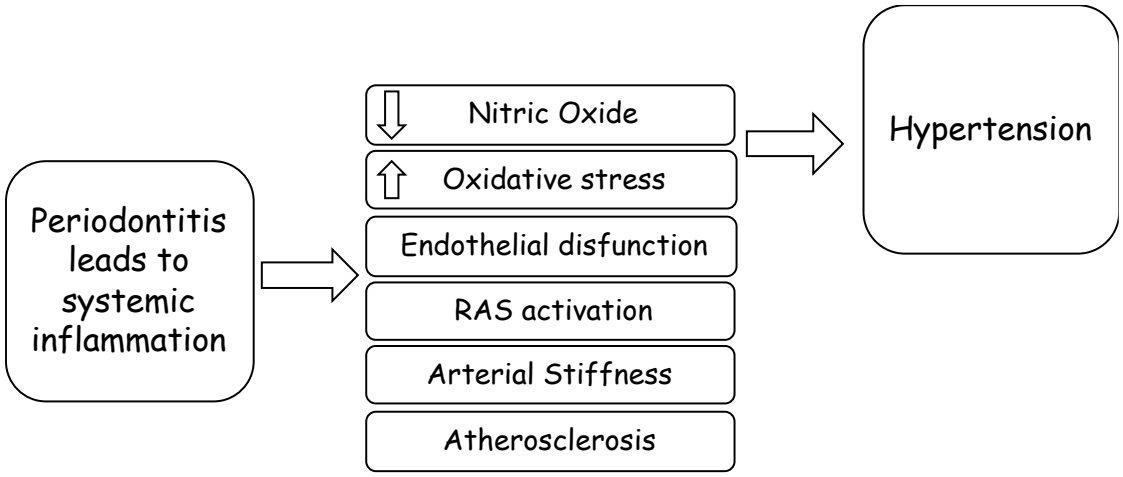


Figure 3. Possible mechanisms between periodontitis and hypertension. Adapted from Macedo-Paizan 2014 (109).

Periodontitis and hypertension are extremely prevalent diseases globally, which have been independently associated to major cardiovascular events, representing a significant disease burden with an impact on health expenditures (8, 110). However, the relationship between these two disorders is not yet well understood and whether this is a simply correlative association or if in fact, the link relates to a causal mechanistic interaction. Additionally, achieving periodontal health could influence blood pressure regulation and incident hypertension. Hence, elucidating the nature of the association between these major health problems is of public health importance and imperative from a medical/therapeutical standpoint. A thorough appraisal of the available evidence linking periodontitis and hypertension is warranted.

Hypothesis and Objectives

HYPOTHESIS AND OBJECTIVES

2.1. OVERALL RESEARCH QUESTION

What is the nature of the relationship between periodontitis and hypertension?

2.2. SPECIFIC QUESTIONS

- Are patients with periodontitis more likely to have hypertension (compared to those with no periodontitis)?
- Is the degree of hypertension influenced by the severity and/or extent of periodontitis? In other words, is there a linear association?
- Is the mean SBP/DBP higher in patients with periodontitis versus those without periodontitis?
- Does periodontal therapy modify the levels of blood pressure?
- Is the level of systemic inflammation (measured as CRP levels and WBC counts) higher in patients with periodontitis and hypertension?
- Does systemic inflammation mediate the association between periodontitis and hypertension?
- What is the level of undetected hypertension in patients with periodontitis (compared to those with no periodontitis)?

This thesis followed the outlines of this PECO:

Population: Individuals ≥ 16 years-old
Exposure: Presence of periodontitis with/without treatment
Comparison: Individuals with no periodontitis
Outcome: Any measure of prevalence and/or levels of hypertension and/or levels of systemic inflammation and/or changes in blood pressure following periodontal therapy

2.3. OBJECTIVES

PRIMARY OBJECTIVES

To test:

- The null hypothesis of no association between periodontitis and prevalence of hypertension
- The null hypothesis of no association between extent and/or severity of periodontitis and mean systolic and diastolic blood pressure
- The null hypothesis of no association between periodontal therapy and modification of the levels of blood pressure
- The null hypothesis of no association between periodontitis, hypertension and levels of systemic inflammation

SECONDARY OBJECTIVES

To report on:

- Possible mechanisms of association between periodontitis and hypertension
- Definitions and classification of hypertension and periodontitis as reported in existing studies
- Traditional confounding factors including risk modifiers (age, gender, smoking, alcohol, socioeconomic status, physical exercise, ethnicity and obesity)

2.4. OUTCOMES

Primary and secondary outcomes will be defined in the methodology section according to each of the three different study designs and research questions.

To achieve our aims, the following methodological objectives and study designs for the PhD programme will be followed:

a) To investigate the association between periodontitis (extent/severity) and hypertension prevalence, mean blood pressure levels, systemic biomarkers and treatment response using research synthesis (systematic review methodology) to formulate a robust and comprehensive summary of current knowledge providing background and context for primary research studies designed to address the hypotheses. A systematic review and meta-analysis will be performed.

b) To investigate a) the degree of association between periodontitis/periodontal lesions and hypertension, a) the degree of association between periodontitis/periodontal lesions and systemic markers of inflammation (i.e., CRP and WBC counts) and c) the potential role of systemic inflammation in mediating this association using a representative sample of the Korean/North American population. A cross-sectional study will be performed.

c) To investigate a) the association of periodontitis (extent/severity) and mean SBP/DBP; b) the level of possible undetected hypertension [defined as SBP/DBP ≥140/90 mm Hg (Williams et al. 2018) or as SBP/DBP ≥130/80 mm Hg (Whelton et al. 2017)] among otherwise systemically healthy participants with and without periodontitis; and c) the potential role of systemic inflammation in mediating this association using a sample of UK adults who were recruited for various research projects at the Eastman Dental Institute. A post-hoc nested case-control study will be performed.

Material and methods & results

3. MATERIAL & METHODS AND RESULTS

The material & methods and results of this PhD encompasses three independent and complementary study designs exploring the association between periodontitis and hypertension: a systematic review and meta-analysis, a cross-sectional study, and a nested case-control study. The methodology of each of these studies followed the detailed research project protocol for this PhD, which was approved by the CED (Comisión Específíca de Doctorado) in May 2019.

Succinctly, the systematic review and meta-analysis of epidemiological and interventional studies intended to provide a general overview on the odds of hypertension among individuals with periodontitis. The cross-sectional study of two large databases was designed to investigate not only the reproducibility/consistency of the systematic review findings but also to assess the mediation effect of systemic markers of inflammation between these two conditions. The third and last study design involved a nested case-control study, which assessed the relationship between periodontitis and blood pressure in systemically healthy individuals.

The specific description of the material & methods and results of each study design is fully described within each published manuscript herein displayed.

3.1. MATERIAL & METHODS AND RESULTS FROM 1ST ORIGINAL PUBLICATION

Reference: Muñoz Aguilera E, Suvan J, Buti J, Czesnikiewicz-Guzik M, Barbosa Ribeiro A, Orlandi M, et al. Periodontitis is associated with hypertension: a systematic review and meta-analysis. Cardiovascular research. 2020;116(1):28-39.

Title: Periodontitis is associated with hypertension: a systematic review and meta-analysis.

Authors: Muñoz Aguilera, E., Suvan, J., Buti, J., Czesnikiewicz-Guzik, M., Barbosa Ribeiro, A., Orlandi, M., Guzik, T.J., Hingorani, A.D., Nart, J. and D'Aiuto, F.

Journal: Cardiovascular Research
Impact factor: 8.168
Journal quartile: Q1

CVR-2019-0517

Periodontitis is associated with Hypertension. A systematic review and Meta-analysis.

Eva Munoz Aguilera[1, 6], Jeanie Suvan[1], Jacopo Buti[1], Marta Czesnikiewicz-Guzik[2,3], Aline Barbosa Ribeiro[2,4], Marco Orlandi[1], Tomasz J Guzik[2,3], Aroon D Hingorani[5], Jose Nart[6], Francesco D'Aiuto[1]

[1]Periodontology Unit and Department of Clinical Research, UCL Eastman Dental Institute, London WC1X 8LD, United Kingdom
[2]Periodontology Group; Glasgow Dental School; and Institute of Cardiovascular and Medical Sciences, University of Glasgow, United Kingdom
[3]Department of Experimental Dentistry and Dental Prophylaxis and Department of Internal and Agricultural Medicine, Jagiellonian University, Krakow, Poland
[4]Department of Physiology, Ribeirao Preto Medical School. University of Sao Paulo. Brazil
[5]Genetic Epidemiology, Institute of Cardiovascular Science, Faculty of Pop Health Sciences, University College London
[6]Department of Periodontology, Faculty of Dentistry, Universitat Internacional de Catalonia, Sant Cugat del Vallès 08195, Barcelona, Spain

Corresponding author:

Prof Francesco D'Aiuto
Periodontology Unit
UCL Eastman Dental Institute
256 Grays Inn Road
London WC1X 8LD

Tel 0203 456 1108
Fax 0203 456 1137
Email: f.daiuto@ucl.ac.uk

Key words: Hypertension, periodontitis, blood pressure, inflammation, periodontal diseases, oral health, periodontal therapy

Word count:

Competing interest statement: "All authors have completed the Unified Competing Interest form (available on request from the corresponding author) and declare: no support from any organisation for the submitted work; no financial relationships with any organisations that might have an interest in the submitted work in the previous three years, no other relationships or activities that could appear to have influenced the submitted work".

Transparency declaration: I, Francesco D'Aiuto affirm that the manuscript is an honest, accurate, and transparent account of the study being reported; that no important aspects of the study have been omitted; and that any discrepancies from the study as planned have been explained.

Details of funding: We would like to acknowledge that contribution of this work was undertaken at UCLH/UCL who received a proportion of funding from the Department of Health's NIHR Biomedical Research Centre funding scheme. TJG is funded by the European Research Council (ERC) InflammaTENSION project.

Abstract

Recent evidence suggests a link between periodontitis (PD) and hypertension, but the nature of this association remain unclear. The overall aim of this review was to critically appraise the evidence linking these two common disorders. Systematic search was conducted for studies published up to December 2018. Prevalence of hypertension in patients with PD (moderate/severe groups) versus those without PD (non-PD) was the primary outcome. Additional outcomes included adjusted mean difference in systolic (SBP) and diastolic (DBP) blood pressure (BP) levels in PD vs non-PD, assessment of biomarkers in PD and hypertension and BP changes after periodontal therapy. From eighty-one studies selected, 40 were included in quantitative meta-analyses. Diagnoses of moderate-severe PD (OR= 1.22; 95% CI, 1.10, 1.35) and severe PD (OR=1.49; 95% CI, 1.09-2.05) were associated with hypertension. Prospective studies confirmed PD diagnosis increased likelihood of hypertension occurrence (OR=1.68; 95% CI, 0.85-3.35). Patients with PD exhibit higher mean SBP (WMD of 4.49 mmHg; 95% CI, 2.88, 6.11) and DBP (2.03 mmHg; 95% CI, 1.25-2.81) when compared to non-PD. Lastly, only 5 out of 12 interventional studies confirmed a reduction in BP following periodontal therapy, ranging from 3-12.5 mmHg of SBP and from 0-10 mmHg of DBP. Periodontitis is associated with increased odds of hypertension (SORT C) and higher SBP/DBP levels. The evidence suggesting that PD therapy could reduce BP is inconclusive. Although additional research is warranted on this association, these results suggest that oral health assessment and management of periodontitis could not only improves oral/overall health and quality of life but also be important in patients with hypertension.

PROSPERO ID: CRD42017081455

Introduction

Hypertension, defined as values ≥140 mmHg Systolic Blood Pressure (SBP) and/or ≥90 mmHg Diastolic Blood Pressure (DBP), is the most prevalent of all cardiovascular diseases (CVDs)(1). Almost 45% of the worldwide population is affected and the estimate increases steeping with age (2). The incidence of adverse cardiovascular (CV) events such as stroke, myocardial infarction, sudden death, heart failure and peripheral artery disease as well as of end-stage renal disease is strongly associated with hypertension (3, 4). According to the world health organisation (WHO) report in 2014, hypertension accounts for 51% of deaths from stroke and 45% of overall CV mortality and this is true at all ages and in all ethnic groups (2). Blood pressure values are an important predictor of cardiovascular risk (5, 6). Despite available treatments, essential hypertension remains poorly controlled with high rates of no treatment and under-treatment (7). Hence, it is still one of the major modifiable risk factor for CVDs that requires urgent management (8). Hypertension is a complex multifactorial disease with no simple mechanism entirely explaining the blood pressure rise (9). Endothelial dysfunction (as manifested by changes in endothelin and nitric oxide), oxidative stress, and inflammation are implicated in the development of hypertension. Despite a prominent role of the immune system has been observed in experimental models (10) and clinical studies (11) studying the onset of hypertension, the exact mechanisms initiating these responses remain unclear (12).

Periodontitis is a chronic multifactorial inflammatory disease caused by a dysbiotic microflora and resulting in progressive destruction of the dental surrounding tissues and leading to tooth loss. It is associated with masticatory dysfunction and negative impact on the patient's quality of life (13). It is estimated that periodontitis affects over 50% of the worldwide population and its severe form is considered the 6th most prevalent disease of humankind (14, 15). Periodontitis is a major public health problem that considerably increases morbidity and costs of oral health care (16, 17). There is consistent observational evidence that periodontitis is associated with an increased risk for future CVDs independent of traditional risk factors such as smoking and obesity (17, 18). The interplay between the bacterial burden and host response is the most plausible biological mechanism linking periodontitis a number of chronic systemic diseases, such as diabetes mellitus, CVDs and neurological diseases such as Alzheimer (17, 19, 20). An ulcerated epithelial lining of the gingival pocket, subsequent to a local inflammatory response to the dental biofilm could amount to a sizeable area in patients with generalized periodontitis (21). These patients often present with systemic inflammation and endothelial dysfunction (22), which improves following successful periodontal treatment (23).

Several studies appear to support a relationship between severe periodontitis and hypertension (24-27). Limited evidence also suggests that successful periodontal treatment could improve arterial blood pressure (28, 29). However, little is still known about the direction and nature of the association between these two conditions. The overall aim was to conduct a robust critical appraisal of the evidence on the relationship between periodontitis and hypertension. Specific research questions were designed based on the following PECO outline: Population: Individuals >16 years old; Exposure: Presence of periodontitis with/without treatment; Comparison: Individuals with no periodontitis; Outcome(s): Any measure of prevalence and/or levels of hypertension and/or changes in blood pressure following periodontal therapy. In this analysis we addressed several key questions:

- Are patients with periodontitis more likely to have hypertension (compared to those without periodontitis)?
- Is the degree of hypertension influenced by the severity and/or extent of periodontitis (linear association)?
- Is the mean SBP/DBP higher in patients with periodontitis versus those without periodontitis?
- Does periodontal therapy modify the levels of blood pressure?

Material and Methods:

The systematic review protocol was registered in PROSPERO on 28/11/2017 with ID: CRD42017081455. A PRISMA statement is attached to follow the reporting of this systematic review (Appendix 1).

Primary and secondary outcomes:

The primary outcome of this systematic review was OR/RR and CI for hypertension in individuals with periodontitis.

The secondary outcomes included prevalence of hypertension in patients with periodontitis vs patients without periodontitis as well as prevalence of periodontitis in patients with or without hypertension; reports of mean SBP/DBP levels in periodontally-healthy and diseased patients; systemic biomarkers associated with periodontitis and hypertension and changes in BP measurements following periodontal therapy.

Inclusion/Exclusion Criteria

To obtain an estimate of the association between periodontitis and hypertension, inclusion criteria were set to be broad and inclusive. Prospective and retrospective studies were included (randomized controlled trials, controlled clinical trials, cohort studies, case-control studies and cross-sectional studies). Eligibility criteria included individuals from age 16 years onward, with periodontitis (chronic and/or aggressive forms) considered as the exposure. Manuscripts including information related to primary and secondary outcomes were included.

Case report, case series and reviews and animal studies were excluded. Individuals under 16 year's old and pregnant women were also excluded. Lastly, studies that did not have any reports of the primary or secondary outcomes were disqualified.

Search methods for identification of studies:

Five electronic databases were searched up to the 10th December 2018 with no year restrictions (Cochrane Central Register of Controlled Trials (CENTRAL), MEDLINE (OVID), EMBASE, Web of science, LILACS). The search included no language restrictions and attempts were made to translate non-English language manuscripts (if this was not possible then the relevant evidence was excluded). In addition, SIGLE database was examined for relevant unpublished trials. Performed detailed search strategies deemed appropriate for each database using a combination of controlled vocabulary (MeSH terms and free text terms). Supplementary table with all terms is available (Appendix 2).

MeSH terms in all trees and subheadings: "periodontal diseases", "periodontics", "hypertension", "blood pressure".

Keywords: "periodont$", "gingiv$", "((blood or bleed$) adj4 prob$)", "(ging$ adj disease)", "hypertens$", "((elevat$ or high$ or rais$) adj3 (diastolic or systolic or arterial or blood) adj pressure)", "bloodpressure".

Hand searching of bibliographies of papers and review articles retrieved articles not found through other search methods.

Data management:

The eligibility assessment of titles and abstracts (when available) of all reports identified were independently screened by two reviewers based on inclusion/exclusion criteria (EMA & JS). If agreement could not be reached, the study was moved to the next stage and inclusion was based on full text screening. Full reports were obtained and assessed independently and in duplicate (EMA and JS) for studies seeming to meet the inclusion criteria or for which insufficient information in the title and abstract precluded to make a clear decision. Disagreements were resolved by discussion and if necessary, a third reviewer was consulted (FD). When authors were not reporting on an effect estimate they were contacted to request additional information. Excel sheets were created to document information regarding decision for included and excluded articles. Kappa statistic was used to assess the agreement between the reviewers based on full text screening.

The main categories of data grouped according to study design and reported in evidence tables were study characteristics data; population; exposure (case definition for periodontitis); outcome (case definition for hypertension); effect (OR/RR with CI); and publication conclusions.

Regarding the exposure, multiple case definitions for periodontitis were found. A lack of consistent case definitions contributed to the difficultly in assessment and interpretation of the data retrieved. In order to collate studies looking at similar definitions, results were therefore grouped using two case definition thresholds: confident and non-confident case definition of periodontitis based on the following criteria (adapted from a previously reported definition) ((30).

Confident case definition of periodontitis: The following criteria were considered as a confident case definition for periodontitis: Generalized chronic periodontitis (at least 30% sites with CAL ≥ 4 mm) (31); at least 2 sites on different teeth with clinical attachment level (CAL) 6 mm and at least 1 site with probing pocket depth (PPD) 4 mm (CDC/AAP periodontitis definition) (32); presence of proximal attachment loss of > 3mm in two or more nonadjacent teeth (sensitive definition) or presence of proximal attachment loss of >5 mm in > 30% of teeth present (33); at least 5 sites with CAL≥6 mm (34).

Non-confident case definition of periodontitis: For non-confident case definition the following reported criteria were considered: Community periodontal index (CPI) score 3/4 in at least 1 quadrant; “Alveolar bone loss” without other measurements of PPD/CAL; unclear diagnostic criteria for periodontitis.

Regarding the outcome, hypertension was defined as SBP ≥ 140 mm Hg / DBP ≥ 90 mm Hg or the use of anti-hypertensive medications (1, 2). However, reports of BP levels and other cases definitions were also documented for, such as self-reported hypertension and other thresholds (high normal/prehypertension).

Assessment of bias individual studies:

Quality assessment of all included studies were undertaken independently and in duplicate by two reviewers as part of the data extraction process. For bias assessment of randomised controlled trials, non-randomised studies of interventions, and observational studies we used the revised Cochrane tool (ROB-2.0 tool) (35), the ROBINS-I tool (36) and the Newcastle-Ottawa (NOS) tool (37) respectively.

Data Synthesis:

Descriptive statistics were performed to summarise the evidence retrieved and to determine the quantity of data, checking further for study variations in terms of study characteristics and results. This assisted in confirming the suitability of further synthesis methods.

Meta-analysis A was conducted and referred to the following primary outcome: ORs for hypertension among people with or without a diagnosis of periodontitis. The ORs with adjustment for the confounding variables (i.e. age, gender, smoking, socioeconomic status, systemic disease, medication, body mass index etc.) was chosen with hypertension as the dependent variable and periodontitis as the independent variable. Pooled estimates of OR and corresponding 95% confidence intervals, were calculated for dichotomous data. In presence of significant heterogeneity (p<0.1), the pooled estimates of effects were calculated using random effects models rather than fixed effects models. Meta-analysis B referred to the secondary outcome (mean SBP/DBP). The pooled mean SBP/DBP difference and 95% confidence intervals were estimated for continuous data. RevMan® 5.3 and JMP® 13.0.0 were used for all the statistical analyses.

To evaluate whether the methodological quality of the included studies influenced the direction or the magnitude of the results, we performed a separate sensitivity analysis by study design and either disease severity or case definition (Fig. 2-Fig. 3C and Appendix 5).

Publication bias:

Possible publication bias was assessed for studies included in the different meta-analyses A and B using the methods described by Begg and Egger (38, 39).

Heterogeneity:

The significance of any discrepancies in the estimates from different trials was assessed by means of Cochran's test for heterogeneity and the I2 statistic. As alluded above, sensitivity analyses were also planned to explore, quantify and control for sources of heterogeneity between studies.

Strength of recommendation:

The quality and strength of the evidence was assessed with The SORT (Strength of Recommendation Taxonomy). The authors discussed the outcomes of the systematic review, pertinent sources of evidence, clinical recommendations and future areas requiring research (40).

Results:

The electronic search from combination of all databases identified 5575 potentially relevant articles after removal of duplicates, resulting in 182 publications eligible for full text screening. Eighty-one publications met inclusion criteria (Figure 1). The evidence tables created according to study design, included the main study characteristics (Appendix 3). The studies included in the systematic review have been conducted in 26 different countries from all continents involving a large variety of different populations. Authors achieved an almost perfect agreement with a percentage of 97.24%; Cohen's k: 0.94.

A variety of case definition of periodontitis were identified (as shown in evidence tables, Appendix 3). For hypertension diagnosis, the studies generally reported more uniformed criteria based on levels of SBP ≥140mm Hg and/or DBP ≥90 mm Hg or use of antihypertensive medication (41). Nevertheless, some studies reported lower cut offs for hypertension (i.e. SBP ≥130mm Hg and/or DBP ≥85 mm Hg) (42) or hypertension prevalence was based on medical records, self-report or national classification codes for disease. Similarly, different methods for measuring blood pressure were described in the studies included (Appendix 3). For additional or missing data, of all the authors contacted, only 3 provided additional information regarding the direction of the association and/or mean SBP/DBP following periodontal therapy (43-45).

Study quality for observational studies as assessed by the Newcastle Ottawa scale varied across the studies, ranging from a score of 3/9 to 9/9 (Appendix 4). The assessment revealed several

potential sources of bias including to the adequacy of case definition for cases and controls, the representativeness of the cases, no appropriate description of the sample size calculation, lack of adjustment for potential confounders or inappropriate statistical test. The assessment of randomise controlled trials with the Rob 2.0 tool revealed a low (5 studies) to high (2 studies) risk of bias for the studies included (Appendix 4). The main reasons for high risk of bias in randomised controlled trials arise from the randomization process, blinding of participants and personnel. Study quality for non-randomized trials revealed moderate and serious risk of bias for the two studies assessed with the Robinson I tool (Appendix 4).

Primary outcome

Twenty studies included in 5 meta-analyses (A) of cohort, cross sectional and case-control studies (Fig. 2-Fig. 3C and Appendix 5) compared the odds of having hypertension if an individual had periodontitis versus periodontally healthy individuals using a periodontal case definition as the exposure measure.

Statistically significant heterogeneity was confirmed with a Tau-squared test (ranging from 0.32 to 0.03), Chi-squared test ranging from (ranging from <0.00001 to 0.008) and I2 test (ranging from 63% to 92%) for the different analyses completed. Due to this level of heterogeneity observed in the studies, random effect meta-analysis was performed.

Odds ratios ranged from 0.90 to 4.20 for all studies, depending on case definition applied, severity of periodontitis and adjustment of the models. Precision of the estimates in the studies varied considerably as appreciated in the varying span of the confidence intervals. Two studies (24, 46) reported ORs for moderate to severe periodontitis separately and one study (47) reported OR for men and women also separately, therefore these different ORs were included independently.

The analysis of three cohort studies predicted the occurrence of hypertension (OR=1.68; 95% CI, 0.85-3.35), but this was not statistically significant (p=0.14) (Appendix 5). Three studies were excluded from this meta-analysis due to one of them reported RR (48) and the other two appeared to be duplicated data (45, 49). Diagnosis of moderate-severe periodontitis in 15 cross-sectional and case-control studies was associated with higher odds of hypertension (1.22, 95% CI 1.10, 1.35), which was statistically significant (p=0.0001) (Fig. 2). A meta-analysis of 8 cross-sectional and case-control studies confirmed that patients with severe periodontitis had increased odds (1.49, 95% CI 1.09, 2.50; p=0.01) of diagnosis of hypertension (Figure 3A). Additionally, meta-analyses of studies according to confident

versus non-confident case definitions of periodontitis were performed. Seven studies with confident definition of periodontitis confirmed higher odds of hypertension (1.53, 95% CI 1.11, 2.10; p=0.009) compared to a meta-analysis of 8 studies with a non-confident definition of periodontitis (1.33, 95% CI 1.14, 1.55; p=0.003) (Figures 3B and 3C).

Secondary outcomes:

Prevalence: Thirty studies reported prevalence of hypertension in patients with periodontitis versus patients without periodontitis or gingivitis (Appendix 6). Twenty-five of these studies showed a higher prevalence of hypertension in patients with a diagnosis of periodontitis (range= 7-77%) versus those without periodontitis (range=4-70%) and one study only confirmed higher prevalence in men (50). These findings were not confirmed in 4 studies (51-54). In addition, a consistent increased prevalence of periodontitis in patients with hypertension (range=29-61%) versus those without hypertension (range=17-39%) was reported in all the 7 publications that included this outcome (Appendix 6).

Mean blood pressure (observational evidence): Thirty-one studies reported average mean SBP/DBP in patients with (range SBP=113-172/DBP=66-101mmHg) and without periodontitis (range SBP=109-143/DBP=65-94mmHg) (Appendix 7). The meta-analysis B, of mean SBP/DBP of 26 studies was performed resulting in statistically significant heterogeneity, confirmed with a Tau-squared test (ranging from 14.38 to 2.92), Chi-squared test ranging from (<0.00001) and I2 test (ranging from 96% to 98%). Patients with periodontitis exhibited higher SBP (WMD of 4.49mmHg, 95% CI 2.88, 6.11; p<0.00001) and DBP (WMD of 2.03mmHg, 95% CI 1.25, 2.81; p<0.00001) when compared to patients without periodontitis (Figures 4 and 5).

Systemic Biomarkers: Three studies were included in the review as reporting changes in systemic biomarkers associated with hypertension and periodontitis (55-57). One study analysed serum levels of neutrophilic enzymes in 95 patients (55). They included a test group of patients with hypertension and periodontitis and 2 control groups: a healthy group (without periodontitis or hypertension) and a hypertensive group. The authors observed that circulating levels of matrix metalloproteinases (MMP)-8, MMP-9, myeloperoxidase and neutrophil elastase (NE) were increased in patients with hypertension and periodontitis but not in the controls. Another study examining the gingival crevicular fluid (GCF) levels in patients with hypertension (21 patients) and without hypertension (26 patients) measuring levels of 8-isoprostane, interleukin (IL)-1B, monocyte chemoattractant protein (MCP)-1, tumour necrosis factor (TNF)α, C-reactive protein (CRP), and MMP-

8 (56). They reported that independent of hypertension present or absent, an increased level of these biomarkers was observed when patients had periodontal pockets. In addition, patients with hypertension presented with almost twice as much periodontal clinical attachment loss (CAL) as controls (Mean + SEM in HTN= 0.87±0.13 vs non-HTN= 0.49±0.11). Albush and co-workers assessed levels of vascular thrombotic markers in 40 hypertensive patients with periodontitis (57). Platelet count, fibrinogen, Von Willebrand factor antigen (vWF:Ag) and D-Dimer levels increased after 48h of treatment (scaling of the teeth including subgingival root debridement for half of the patients and surgical periodontal therapy for the other 20) and decreased after 6 weeks ($p<0.05$), with no significant differences between groups ($p>0.05$). Acute increase in endothelial-activation markers including E-selectin, vWF, haemoglobin and haematocrit, D-dimer levels and neutrophils counts was also reported 24 hours following periodontal therapy in several publications (22, 44, 58). Reductions in inflammatory biomarkers were observed in 11 interventional studies following periodontal therapy (22, 28, 29, 44, 59-65).

Mean Blood Pressure (interventional evidence): The search located 12 interventional clinical trials reporting the effect of periodontal therapy on blood pressure either as a primary [(65)] or secondary outcome (the remaining 11 studies) (See Appendix 3 for a detailed description of the studies and treatment modalities). Eight studies were RCTs, 3 were non-RCT and 1 was a pilot study. These studies comprised a varied sample of individuals, including people medically healthy in 6 studies (22, 28, 44, 58, 59, 62) pre-hypertension (65), refractory hypertension (29), hypertension (59), metabolic syndrome (63), coronary artery disease (60) and type 2 diabetes (64). Five of the 12 interventional studies included in the analysis confirmed a reduction in SBP following periodontal therapy (range=3-12.5 mmHg), and an inconsistent reduction of DBP (range=0-10 mmHg) (28, 29, 61, 64, 65) . Six studies reported no changes in blood pressure measures following non-surgical and/or surgical periodontal therapy (44, 58-60, 62, 63), however, only 2 studies reported actual blood pressure values (59, 60) and one author provided values upon request (44). One study (22) reported an increase in blood pressure in the test group one day after periodontal therapy.

Publication bias:

Study publication bias was examined using funnel plots for both meta-analyses A and B (Appendix 8). Egger's test was only calculated for the meta-analyses A of moderate to severe periodontitis (Appendix 8). Visual assessment of the Funnel of moderate-to severe periodontitis revealed studies were slightly skewed to the right, which was confirmed with the Egger's test showing a statistically significant difference ($p=0.0054$); publication bias was therefore suspected in this

analysis. Nevertheless, all the other funnel plots for meta-analyses A displayed symmetrical appearance. Similarly, visual assessment of the Funnel plots for mean SBP and DBP analysis revealed symmetrical appearance. Egger's test estimated for meta-analyses B revealed a not statistically significant result (p=0.5582) for the mean SBP meta-analysis and a statistically significant difference (p=0.0224) for the mean DBP meta-analysis. On this basis, publication bias was suspected in the mean DBP meta-analysis.

Reporting on strength of recommendation:

The quality, quantity and consistency of the evidence from observational and interventional studies on the relationship between periodontitis and hypertension was thoroughly assessed for a SORT recommendation. Accordingly, we conclude that diagnosis and treatment of periodontitis is positively associated with hypertension (SORT C) (40).

Discussion:

The results of this systematic review support a positive association between periodontitis and hypertension. Based on the quantitative analyses of all studies included, patients with moderate to severe periodontitis have greater (20%) odds of having hypertension when compared to patients without periodontitis. In addition, a positive linear association was observed, confirming that the more severe periodontitis is, the higher the likelihood (49%) of having hypertension. This finding was further corroborated, when the studies with a confident case definition for periodontitis were analysed, confirming even greater odds (50%) of diagnosis of hypertension were found. The magnitude of association between periodontitis and hypertension reported in this review (OR 1.22 to 1.53) is in agreement with that recently reported (27). In this recent review, however Martin-Cabezas and co-workers included observational studies without specifying the exposure and outcome of the analysis. In the current systematic review, we also included three cohort studies (66-68) confirming a temporal association between periodontitis and incidence of hypertension although this was not statistically significant and we excluded a number of studies in this analysis in order to avoid bias due to duplication of data was suspected.

This systematic review also confirmed an increased prevalence of periodontitis in patients with hypertension (as defined by SBP≥ 140 and DBP≥ 90 mm Hg). Clinical and experimental evidence suggest that this direction of the association could be mediated through hypertension causing microcirculatory changes in of the gingival tissue leading to ischemia, increased inflammation and/or altered microbial composition of the dental biofilm (25, 69, 70). This finding combined with the

increased prevalence of hypertension in patients with periodontitis could be even more significant within the context of the new revised guidelines issued by the AHA for the definition of hypertension (71). A reduced threshold of SBP/DBP for the case definition of hypertension was expressed (stage 1 as SBP=130–139/DBP=80–89 mm Hg, and stage 2 agreeing to stages 1 and 2 in the JNC 7 report; i.e. SBP≥140/DBP≥90 mm Hg), which has been reported in a recent cross-sectional study to double the prevalence estimates of hypertension in countries like China and US (72). This could result in even greater odds of hypertension in patients with periodontitis and vice versa. Future research should consider the impact of these thresholds for case definition of hypertension in terms of increased prevalence and treatment thereof.

In this systematic review, for the first time, we attempted to provide an estimate of the mean arterial BP in patients with periodontitis versus controls. Very interestingly, more than 80% of the included studies reporting levels of blood pressure showed consistently increased levels of systolic and diastolic BP in patients with periodontitis. Further, the exploratory meta-analysis B revealed that patients with periodontitis showed a higher MWD of 4.49mmHg of SBP and of 2.03 mmHg of DBP. If confirmed in long-term longitudinal studies, periodontitis could represent a novel modifiable risk factor for hypertension at the same strength of diabetes and smoking (73, 74). However, as periodontitis, diabetes and hypertension share common risk factors (such as aging, smoking, and disadvantageous socioeconomic status, among others), residual confounding could affect the magnitude of these associations. It is important to state that this association could also be driven by an association between arterial blood pressure changes and other undetected sources/chronic infections. Further research in identifying the interplay between triggers/bacterial burdens in each individual and their relative contribution on blood pressure is needed.

Raised arterial blood pressure observed in periodontitis could also explain the moderate but consistent higher risk of CV events (i.e. MI and stroke) reported by several investigators in patients with periodontitis when compared to controls (17). Indeed, an average increase of 5 mm Hg of SBP has been consistently associated with a 25% increased mortality from ischemic heart disease and stroke (75). These assumptions should all be interpreted with caution because of the high heterogeneity observed in the reported scientific evidence. In particular, varying case definitions of periodontitis and hypertension could have undermined the validity of these observations. Nevertheless, due to the high prevalence of both conditions, the clinical implications for public health systems could be very significant.

This systematic review also confirmed a potential positive effect of treating periodontal inflammation on arterial blood pressure. Inconclusive findings were identified in the selected studies, with only 5 out of 12 intervention trials showing a reduction of SBP/DBP in patients with periodontitis. Only one RCT was designed to address the question whether non-surgical periodontal therapy could result in reduced arterial BP levels (65). These authors assessed changes in blood pressure as their primary outcome following non-surgical periodontal therapy. They included 107 pre-hypertensive participants and reported an absolute difference of SBP=12.57mmHg 95% CI 10.45-14.69, $p<0.05$ and of DBP=9.65mmHg 95% CI 7.06-12.24, $p<0.05$ after periodontal therapy. As treatment of hypertension has been repeatedly advocated as a key intervention to improve general health, quality of life and reduce CV complications (76), periodontitis treatment could represent a novel non-pharmacological therapy to prevent/help manage hypertension. A meta-analysis of RCTs quantified a reduction of 25-30% of coronary heart disease events such as stroke and heart failure with a 10-mmHg reduction in SBP or a 5-mmHg reduction in DBP following antihypertensive drug therapy (77). Future research should address the hypothesis of the treatment of periodontitis could achieve similar reduction in arterial BP and CV outcomes.

The identification of periodontitis as a possible risk factor for hypertension could be explained by a number of plausible mechanisms (Figure 6). Firstly, periodontitis is associated with systemic inflammation, mediators of which, including CRP, IL-6; TNF-a can all affect endothelial function. Clinical evidence suggests periodontitis affects systemic endothelial function and in turn this could impact on hypertension. Our group previously demonstrated that treatment of severe periodontitis improves endothelial function by reduction in systemic inflammation in patients with and without other comorbidities like diabetes (22, 64). Secondly, some reports suggest possible direct effects of oral microbiota related bacteraemia in mediating vascular dysfunction as well. Emerging experimental animal evidence indicates that an immune response to a common periodontal pathogen: *Porphyromonas gingivalis* (*Pg*) results in elevation of BP, vascular inflammation and endothelial dysfunction (78). Another possibility may be that cells, including T cells, B cells as well as monocyte/macrophages, primed in inflamed periodontium may be more prone to chemotactic recruitment to perivascular adipose tissue and adventitia, a step that has been shown to precede development of vascular dysfunction, hypertension and atherosclerosis (79, 80). This review therefore raises an important question regarding the causal nature of the association between periodontitis and hypertension.

Strengths and weaknesses

This systematic review was designed to comprehensively investigate the possible role of periodontitis as a possible novel risk factor for hypertension. A number of limitations however should be highlighted starting with the limited value of systematic reviews of observational studies for ascertaining causality (81). Moreover, observational studies have intrinsic biases (mostly selection and information bias), hence the results of this systematic review should be interpreted within the context of the methodology used. Nevertheless, this review was broad and inclusive of not only observational but also interventional studies. Besides, because of the link between periodontitis and cardio-metabolic risk factors (17, 82), this review also included data from observational studies on MetS and CVD but the authors acknowledge that some of the data may have been missed due to the difficulties in identifying the outcomes within the published reports. Moreover, studies looking at hypertension have inherent problem of the effect of blood pressure measurement technique on the outcome as well as variable degree of reporting of actual criteria of hypertension. Therefore, we have focused on a clear definition of hypertension basing mainly on blood pressure values and anti-hypertensive medications. With the exception of a single study (29) most studies have used office rather than ambulatory blood pressure; our quantitative analysis of the effect of periodontitis on blood pressure values adds to the strength of the selected evidence. This study was a pilot intervention trial including only 26 patients with refractory hypertension and periodontitis and the effects of non-surgical periodontal therapy on both systolic and diastolic blood pressure were of greater magnitude of those reported in the other intervention studies. We urge caution in interpreting these results especially in view of the limited sample size and inclusion criteria adopted by the authors (29). Future intervention trials should all be powered and include ambulatory blood pressure levels.

One of challenges encountered was to establish the direction of association when studies were included in the quantitative analyses (i.e. dependent and independent variables in the model). This was mainly due to unclear description in the published manuscripts. When a consensus could not be achieved among the reviewers (EMA, JS), a third author was consulted (FD) or attempts were made to contact the authors for clarification (43). Another important limitation of this systematic review is the high level of heterogeneity in the case definitions for both, periodontitis and hypertension (34, 83). To overcome this, data was further analysed according to an arbitrary level of confidence in a given case definition of periodontitis. In fact, when an arbitrary confident diagnosis was confirmed, the observed magnitude of association between periodontitis and hypertension was greater. The lack of consistent measures of case-definition and severity of periodontitis in the retrieved evidence did not allow for a relevant analysis of extent and severity of periodontitis with all endpoints of blood pressure. We hope in the future reporting of periodontitis is more consistent and allow for such analyses. Lastly, it has been reported that antihypertensives such as calcium channel blockers can

cause gingival enlargement in 6.3% to 83% of patients (Hallmon & Rossmann 1999), which should not be confounded with periodontitis.

Conclusions:

Periodontitis could be associated with increased risk of hypertension in a linear fashion. Further, management of periodontitis could impact on the management of hypertension. Our findings highlight the potential to improve CV outcomes by addressing poor oral health in the general population. Longer and larger studies are needed however to determine whether periodontal treatment benefit patients in terms of CV health, ultimately resulting in reduced morbidity and mortality.

Translational Implications:

- To raise awareness of the association between periodontitis and hypertension.
- Patients with periodontitis should be informed by oral health professionals of the risk of developing hypertension.
- Oral health advice should be given to all patients with hypertension.
- Prevention and management of periodontitis improves oral/overall health and quality of life and could prevent/improve hypertension.
- Larger observational studies should include internationally recognised case definitions for periodontitis and hypertension.
- More RCT with reduction of blood pressure as the primary outcome.
- Reporting of patient related outcome measures within the study designs.

Acknowledgement

We would like to acknowledge that contribution of this work was undertaken at UCLH/UCL who received a proportion of funding from the Department of Health's NIHR Biomedical Research Centre funding scheme. TJG is funded by European Research Council (ERC) InflammaTENSION project.

References

1. Williams B, Mancia G, Spiering W, Agabiti Rosei E, Azizi M, Burnier M, Clement DL, Coca A, de Simone G, Dominiczak A, Kahan T, Mahfoud F, Redon J, Ruilope L, Zanchetti A, Kerins M, Kjeldsen SE, Kreutz R, Laurent S, Lip GYH, McManus R, Narkiewicz K, Ruschitzka F, Schmieder RE, Shlyakhto E, Tsioufis C, Aboyans V, Desormais I, Authors/Task Force M. 2018 ESC/ESH Guidelines for the management of arterial hypertension: The Task Force for the management of arterial hypertension of the European Society of Cardiology and the European Society of Hypertension: The Task Force for the management of arterial hypertension of the European Society of Cardiology and the European Society of Hypertension. *J Hypertens* 2018;**36**:1953-2041.

2. Mancia G, Fagard R, Narkiewicz K, Redon J, Zanchetti A, Böhm M, Christiaens T, Cifkova R, De Backer G, Dominiczak A. 2013 ESH/ESC guidelines for the management of arterial hypertension: the Task Force for the Management of Arterial Hypertension of the European Society of Hypertension (ESH) and of the European Society of Cardiology (ESC). *Blood pressure* 2013;**22**:193-278.

3. Bath P, Chalmers J, Powers W, Beilin L, Davis S, Lenfant C, Mancia G, Neal B, Whitworth J, Zanchetti A. International Society of Hypertension (ISH): statement on the management of blood pressure in acute stroke. *Journal of hypertension* 2003;**21**:665.

4. Mancia G, Laurent S, Agabiti-Rosei E, Ambrosioni E, Burnier M, Caulfield MJ, Cifkova R, Clément D, Coca A, Dominiczak A. Reappraisal of European guidelines on hypertension management: a European Society of Hypertension Task Force document. *Blood pressure* 2009;**18**:308-347.

5. Bangalore S, Toklu B, Gianos E, Schwartzbard A, Weintraub H, Ogedegbe G, Messerli FH. Optimal systolic blood pressure target after SPRINT: insights from a network meta-analysis of randomized trials. *The American journal of medicine* 2017;**130**:707-719. e708.

6. Bundy JD, Li C, Stuchlik P, Bu X, Kelly TN, Mills KT, He H, Chen J, Whelton PK, He J. Systolic blood pressure reduction and risk of cardiovascular disease and mortality: a systematic review and network meta-analysis. *JAMA cardiology* 2017;**2**:775-781.

7. Joffres M, Falaschetti E, Gillespie C, Robitaille C, Loustalot F, Poulter N, McAlister FA, Johansen H, Baclic O, Campbell N. Hypertension prevalence, awareness, treatment and control in national surveys from England, the USA and Canada, and correlation with stroke and ischaemic heart disease mortality: a cross-sectional study. *BMJ open* 2013;**3**:e003423.

8. Carretero OA, Oparil S. Essential hypertension part I: definition and etiology. *Circulation* 2000;**101**:329-335.

9. Czopek A, Moorhouse R, Guyonnet L, Farrah T, Lenoir O, Owen E, van Bragt J, Costello HM, Menolascina F, Baudrie V. A novel role for myeloid endothelin-B receptors in hypertension. *European heart journal* 2019.

10. Guzik TJ, Hoch NE, Brown KA, McCann LA, Rahman A, Dikalov S, Goronzy J, Weyand C, Harrison DG. Role of the T cell in the genesis of angiotensin II–induced hypertension and vascular dysfunction. *Journal of Experimental Medicine* 2007;**204**:2449-2460.

11. Itani HA, McMaster Jr WG, Saleh MA, Nazarewicz RR, Mikolajczyk TP, Kaszuba AM, Konior A, Prejbisz A, Januszewicz A, Norlander AE. Activation of human T cells in hypertension: studies of humanized mice and hypertensive humans. *Hypertension* 2016;**68**:123-132.

12. Drummond GR, Vinh A, Guzik TJ, Sobey CG. Immune mechanisms of hypertension. *Nature Reviews Immunology* 2019:1.

13. Buset S, Walter C, Friedmann A, Weiger R, Borgnakke WS, Zitzmann NU. Are periodontal diseases really silent? A systematic review of their effect on quality of life. *Journal of clinical periodontology* 2016.

14. Sanz M, D'Aiuto F, Deanfield J, Fernandez-Avilés F. European workshop in periodontal health and cardiovascular disease—scientific evidence on the association between periodontal and cardiovascular diseases: a review of the literature. *European Heart Journal Supplements* 2010;**12**:B3-B12.

15. Kassebaum NJ, Bernabé E, Dahiya M, Bhandari B, Murray CJL, Marcenes W. Global Burden of Severe Periodontitis in 1990-2010 A Systematic Review and Meta-regression. *Journal of dental research* 2014:0022034514552491.

16. Baehni P, Tonetti MS. Conclusions and consensus statements on periodontal health, policy and education in Europe: a call for action–consensus view 1. *European Journal of Dental Education* 2010;**14**:2-3.

17. Tonetti MS, Dyke TE. Periodontitis and atherosclerotic cardiovascular disease: consensus report of the Joint EFP/AAP Workshop on Periodontitis and Systemic Diseases. *Journal of clinical periodontology* 2013;**40**.

18. Demmer RT, Papapanou PN. Epidemiologic patterns of chronic and aggressive periodontitis. *Periodontology 2000* 2010;**53**:28-44.

19. Southerland JH. Periodontitis may contribute to poor control of hypertension in older adults. *Journal of Evidence Based Dental Practice* 2013;**13**:125-127.

20. Dominy SS, Lynch C, Ermini F, Benedyk M, Marczyk A, Konradi A, Nguyen M, Haditsch U, Raha D, Griffin C. Porphyromonas gingivalis in Alzheimer's disease brains: Evidence for disease causation and treatment with small-molecule inhibitors. *Science Advances* 2019;**5**:eaau3333.

21. Hujoel PP, Drangsholt M, Spiekerman C, Derouen TA. Examining the link between coronary heart disease and the elimination of chronic dental infections. *The Journal of the American Dental Association* 2001;**132**:883-889.

22. Tonetti MS, D'aiuto F, Nibali L, Donald A, Storry C, Parkar M, Suvan J, Hingorani AD, Vallance P, Deanfield J. Treatment of periodontitis and endothelial function. *New England Journal of Medicine* 2007;**356**:911-920.

23. Orlandi M, Suvan J, Petrie A, Donos N, Masi S, Hingorani A, Deanfield J, D'Aiuto F. Association between periodontal disease and its treatment, flow-mediated dilatation and carotid intima-media thickness: a systematic review and meta-analysis. *Atherosclerosis* 2014;**236**:39-46.

24. Tsakos G, Sabbah W, Hingorani AD, Netuveli G, Donos N, Watt RG, D'Aiuto F. Is periodontal inflammation associated with raised blood pressure? Evidence from a National US survey. *Journal of hypertension* 2010;**28**:2386-2393.

25. Tsioufis C, Kasiakogias A, Thomopoulos C, Stefanadis C. Periodontitis and blood pressure: the concept of dental hypertension. *Atherosclerosis* 2011;**219**:1-9.

26. Zeigler CC, Wondimu B, Marcus C, Modéer T. Pathological periodontal pockets are associated with raised diastolic blood pressure in obese adolescents. *BMC oral health* 2015;**15**:1.

27. Martin-Cabezas R, Seelam N, Petit C, Agossa K, Gaertner S, Tenenbaum H, Davideau J-L, Huck O. Association between periodontitis and arterial hypertension: A systematic review and meta-analysis. *American heart journal* 2016;**180**:98-112.

28. D'Aiuto F, Parkar M, Nibali L, Suvan J, Lessem J, Tonetti MS. Periodontal infections cause changes in traditional and novel cardiovascular risk factors: results from a randomized controlled clinical trial. *American heart journal* 2006;**151**:977-984.

29. Vidal F, Cordovil I, Figueredo CMS, Fischer RG. Non-surgical periodontal treatment reduces cardiovascular risk in refractory hypertensive patients: a pilot study. *Journal of clinical periodontology* 2013;**40**:681-687.

30. Nibali L, Farias BC, Vajgel A, Tu Y, Donos N. Tooth loss in aggressive periodontitis: a systematic review. *Journal of dental research* 2013;**92**:868-875.

31. Armitage GC. Development of a classification system for periodontal diseases and conditions. *Annals of periodontology / the American Academy of Periodontology* 1999;**4**:1-6.

32. Page RC, Eke PI. Case definitions for use in population-based surveillance of periodontitis. *Journal of periodontology* 2007;**78**:1387-1399.

33. Tonetti M, Claffey N, C EWiPg. Advances in the progression of periodontitis and proposal of definitions of a periodontitis case and disease progression for use in risk factor research: Group C Consensus report of the 5th European workshop in periodontology. *Journal of clinical periodontology* 2005;**32**:210-213.

34. Preshaw PM. Definitions of periodontal disease in research. *Journal of Clinical Periodontology* 2009;**36**:1-2.

35. Higgins J, Sterne J, Savović J, Page M, Hróbjartsson A, Boutron I, Reeves B, Eldridge S. A revised tool for assessing risk of bias in randomized trials. *Cochrane Database Syst Rev* 2016;**10**:29-31.

36. Sterne JA, Hernán MA, Reeves BC, Savović J, Berkman ND, Viswanathan M, Henry D, Altman DG, Ansari MT, Boutron I. ROBINS-I: a tool for assessing risk of bias in non-randomised studies of interventions. *Bmj* 2016;**355**:i4919.

37. Wells G, Shea B, O'connell D, Peterson J, Welch V, Losos M, Tugwell P. The Newcastle-Ottawa Scale (NOS) for assessing the quality of nonrandomised studies in meta-analyses. Ottawa (ON): Ottawa Hospital Research Institute; 2009. *Available in March* 2016.

38. Begg CB, Mazumdar M. Operating characteristics of a rank correlation test for publication bias. *Biometrics* 1994:1088-1101.

39. Egger M, Smith GD, Schneider M, Minder C. Bias in meta-analysis detected by a simple, graphical test. *Bmj* 1997;**315**:629-634.

40. Newman MG, Weyant R, Hujoel P. JEBDP improves grading system and adopts strength of recommendation taxonomy grading (SORT) for guidelines and systematic reviews. *Journal of Evidence Based Dental Practice* 2007;**7**:147-150.

41. Chobanian AV, Bakris GL, Black HR, Cushman WC, Green LA, Izzo Jr JL, Jones DW, Materson BJ, Oparil S, Wright Jr JT. The seventh report of the joint national committee on prevention, detection, evaluation, and treatment of high blood pressure: the JNC 7 report. *Jama* 2003;**289**:2560-2571.

42. Grundy SM, Cleeman JI, Daniels SR, Donato KA, Eckel RH, Franklin BA, Gordon DJ, Krauss RM, Savage PJ, Smith Jr SC. Diagnosis and management of the metabolic syndrome: an American Heart Association/National Heart, Lung, and Blood Institute scientific statement. *Circulation* 2005;**112**:2735-2752.

43. Boland MR, Hripcsak G, Albers DJ, Wei Y, Wilcox AB, Wei J, Li J, Lin S, Breene M, Myers R. Discovering medical conditions associated with periodontitis using linked electronic health records. *Journal of clinical periodontology* 2013;**40**:474-482.

44. Taylor B, Tofler G, Morel-Kopp MC, Carey H, Carter T, Elliott M, Dailey C, Villata L, Ward C, Woodward M. The effect of initial treatment of periodontitis on systemic markers of inflammation and cardiovascular risk: a randomized controlled trial. *European journal of oral sciences* 2010;**118**:350-356.

45. Lee J-H, Lee J-S, Park J-Y, Choi J-K, Kim D-W, Kim Y-T, Choi S-H. Association of lifestyle-related comorbidities with periodontitis: a nationwide cohort study in Korea. *Medicine* 2015;**94**.

46. D'Aiuto F, Sabbah W, Netuveli G, Donos N, Hingorani AD, Deanfield J, Tsakos G. Association of the metabolic syndrome with severe periodontitis in a large US population-based survey. *The Journal of Clinical Endocrinology & Metabolism* 2008;**93**:3989-3994.

47. Tu YK, D'aiuto F, Lin HJ, Chen YW, Chien KL. Relationship between metabolic syndrome and diagnoses of periodontal diseases among participants in a large Taiwanese cohort. *Journal of clinical periodontology* 2013;**40**:994-1000.

48. Rivas-Tumanyan S, Spiegelman D, Curhan GC, Forman JP, Joshipura KJ. Periodontal disease and incidence of hypertension in the health professionals follow-up study. Oxford University Press, 2012.

49. Morita T, Yamazaki Y, Mita A, Takada K, Seto M, Nishinoue N, Sasaki Y, Motohashi M, Maeno M. A cohort study on the association between periodontal disease and the development of metabolic syndrome. *Journal of periodontology* 2010;**81**:512-519.

50. Tuominen R, Reunanen A, Paunio M, Paunio I, Aromaa A. Oral health indicators poorly predict coronary heart disease deaths. *Journal of dental research* 2003;**82**:713-718.

51. Desvarieux M, Demmer RT, Jacobs Jr DR, Rundek T, Boden-Albala B, Sacco RL, Papapanou PN. Periodontal bacteria and hypertension: the oral infections and vascular disease epidemiology study (INVEST). *Journal of hypertension* 2010;**28**:1413.

52. Gomes-Filho IS, das Merces MC, de Santana Passos-Soares J, Seixas da Cruz S, Teixeira Ladeia AM, Trindade SC, de Moraes Marcilio Cerqueira E, Freitas Coelho JM, Marques Monteiro FM, Barreto ML, Pereira Vianna MI, Nascimento Costa Md.a C, Seymour GJ, Scannapieco FA. Severity of Periodontitis and Metabolic Syndrome: Is There an Association? *Journal of periodontology* 2016;**87**:357-366.

53. Han DH, Lim SY, Sun BC, Paek D, Kim HD. The association of metabolic syndrome with periodontal disease is confounded by age and smoking in a korean population: The Shiwha–Banwol environmental health study. *Journal of clinical periodontology* 2010;**37**:609-616.

54. Gordon JH, LaMonte MJ, Genco RJ, Zhao J, Cimato TR, Hovey KM, Wactawski-Wende J. Association of clinical measures of periodontal disease with blood pressure and hypertension among postmenopausal women. *Journal of periodontology* 2018;**89**:1193-1202.

55. Türkoğlu O, Barış N, Tervahartiala T, Şenarslan Ö, Sorsa T, Atilla G. Evaluation of systemic levels of neutrophilic enzymes in patients with hypertension and chronic periodontitis. *Journal of periodontology* 2014;**85**:908-916.

56. Khocht A, Rogers T, Janal M, Brown M. Gingival fluid inflammatory biomarkers and hypertension in African Americans. *JDR Clinical & Translational Research* 2017;**2**:269-277.

57. Albush MM, Razan KK, Al Dieri MR. Effect of surgical and non-surgical periodontal debridement on vascular thrombotic markers in hypertensives. *Journal of Indian Society of Periodontology* 2013;**17**:324.

58. Graziani F, Cei S, Tonetti M, Paolantonio M, Serio R, Sammartino G, Gabriele M, D'aiuto F. Systemic inflammation following non-surgical and surgical periodontal therapy. *Journal of clinical periodontology* 2010;**37**:848-854.

59. Higashi Y, Goto C, Jitsuiki D, Umemura T, Nishioka K, Hidaka T, Takemoto H, Nakamura S, Soga J, Chayama K. Periodontal infection is associated with endothelial dysfunction in healthy subjects and hypertensive patients. *Hypertension* 2008;**51**:446-453.

60. Higashi Y, Goto C, Hidaka T, Soga J, Nakamura S, Fujii Y, Hata T, Idei N, Fujimura N, Chayama K. Oral infection-inflammatory pathway, periodontitis, is a risk factor for endothelial dysfunction in patients with coronary artery disease. *Atherosclerosis* 2009;**206**:604-610.

61. Houcken W, Teeuw WJ, Bizzarro S, Rodriguez EA, Mulders TA, van den Born BJH, Loos BG. Arterial stiffness in periodontitis patients and controls A case-control and pilot intervention study. *Journal of Human Hypertension* 2016;**30**:24-29.

62. Seinost G, Wimmer G, Skerget M, Thaller E, Brodmann M, Gasser R, Bratschko RO, Pilger E. Periodontal treatment improves endothelial dysfunction in patients with severe periodontitis. *American heart journal* 2005;**149**:1050-1054.

63. López NJ, Quintero A, Casanova PA, Ibieta CI, Baelum V, López R. Effects of periodontal therapy on systemic markers of inflammation in patients with metabolic syndrome: a controlled clinical trial. *Journal of periodontology* 2012;**83**:267-278.

64. D'Aiuto F, Gkranias N, Bhowruth D, Khan T, Orlandi M, Suvan J, Masi S, Tsakos G, Hurel S, Hingorani AD. Systemic effects of periodontitis treatment in patients with type 2 diabetes: a 12 month, single-centre, investigator-masked, randomised trial. *The Lancet Diabetes & Endocrinology* 2018;**6**:954-965.

65. Zhou Q-B, Xia W-H, Ren J, Yu B-B, Tong X-Z, Chen Y-B, Chen S, Feng L, Dai J, Tao J, Yang J-Y. Effect of Intensive Periodontal Therapy on Blood Pressure and Endothelial Microparticles in Patients With Prehypertension and Periodontitis: A Randomized Controlled Trial. *Journal of Periodontology* 2017;**88**:711-722.

66. Kawabata Y, Ekuni D, Miyai H, Kataoka K, Yamane M, Mizutani S, Irie K, Azuma T, Tomofuji T, Iwasaki Y, Morita M. Relationship Between Prehypertension/Hypertension and Periodontal Disease: A Prospective Cohort Study. *Am J Hypertens* 2016;**29**:388-396.

67. Morita T, Yamazaki Y, Fujiharu C, Ishii T, Seto M, Nishinoue N, Sasaki Y, Nakai K, Tanaka H, Kawato T. Association between the duration of periodontitis and increased cardiometabolic risk factors: a 9-year cohort study. *Metabolic syndrome and related disorders* 2016;**14**:475-482.

68. Lee J-H, Oh J-Y, Youk T-M, Jeong S-N, Kim Y-T, Choi S-H. Association between periodontal disease and non-communicable diseases: A 12-year longitudinal health-examinee cohort study in South Korea. *Medicine* 2017;**96**.

69. Bonato C, do-Amaral C, Belini L, Salzedas LMP, Oliveira SHPd. Hypertension favors the inflammatory process in rats with experimentally induced periodontitis. *Journal of periodontal research* 2012;**47**:783-792.

70. Demmer RT, Papapanou PN, Jacobs Jr DR, Desvarieux M. Bleeding on probing differentially relates to bacterial profiles: the Oral Infections and Vascular Disease Epidemiology Study. *Journal of clinical periodontology* 2008;**35**:479-486.

71. Whelton PK, Carey RM, Aronow WS, Casey DE, Collins KJ, Himmelfarb CD, DePalma SM, Gidding S, Jamerson KA, Jones DW. 2017 ACC/AHA/AAPA/ABC/ACPM/AGS/APhA/ASH/ASPC/NMA/PCNA guideline for the prevention, detection, evaluation, and management of high blood pressure in adults: a report of the American College of Cardiology/American Heart Association Task Force on Clinical Practice Guidelines. *Journal of the American College of Cardiology* 2018;**71**:e127-e248.

72. Khera R, Lu Y, Lu J, Saxena A, Nasir K, Jiang L, Krumholz HM. Impact of 2017 ACC/AHA guidelines on prevalence of hypertension and eligibility for antihypertensive treatment in United States and China: nationally representative cross sectional study. *bmj* 2018;**362**:k2357.

73. Cheung BM, Li C. Diabetes and hypertension: is there a common metabolic pathway? *Current atherosclerosis reports* 2012;**14**:160-166.

74. Saladini F, Benetti E, Fania C, Mos L, Casiglia E, Palatini P. Effects of smoking on central blood pressure and pressure amplification in hypertension of the young. *Vascular Medicine* 2016;**21**:422-428.

75. Lewington S. Prospective studies collaboration. Age-specific relevance of usual blood pressure to vascular mortality: a meta-analysis of individual data for one million adults in 61 prospective studies. *Lancet* 2002;**360**:1903-1913.

76. Ettehad D, Emdin CA, Kiran A, Anderson SG, Callender T, Emberson J, Chalmers J, Rodgers A, Rahimi K. Blood pressure lowering for prevention of cardiovascular disease and death: a systematic review and meta-analysis. *The Lancet* 2016;**387**:957-967.

77. Law M, Morris J, Wald N. Use of blood pressure lowering drugs in the prevention of cardiovascular disease: meta-analysis of 147 randomised trials in the context of expectations from prospective epidemiological studies. *Bmj* 2009;**338**:b1665.

78. Czesnikiewicz-Guzik M, Nosalski R, Mikolajczyk TP, Vidler F, Dohnal T, Dembowska E, Graham D, Harrison DG, Guzik TJ. Th1-type immune responses to Porphyromonas gingivalis antigens exacerbate angiotensin II-dependent hypertension and vascular dysfunction. *British journal of pharmacology* 2018.

79. Guzik TJ, Skiba DS, Touyz RM, Harrison DG. The role of infiltrating immune cells in dysfunctional adipose tissue. *Cardiovascular research* 2017;**113**:1009-1023.

80. Mikolajczyk TP, Nosalski R, Szczepaniak P, Budzyn K, Osmenda G, Skiba D, Sagan A, Wu J, Vinh A, Marvar PJ. Role of chemokine RANTES in the regulation of perivascular inflammation, T-cell accumulation, and vascular dysfunction in hypertension. *The FASEB Journal* 2016;**30**:1987-1999.

81. Grimes DA, Schulz KF. An overview of clinical research: the lay of the land. *The lancet* 2002;**359**:57-61.

82. Nibali L, Tatarakis N, Needleman I, Tu Y-K, D'aiuto F, Rizzo M, Donos N. Association between metabolic syndrome and periodontitis: a systematic review and meta-analysis. *The Journal of Clinical Endocrinology & Metabolism* 2013;**98**:913-920.

83. Savage A, Eaton KA, Moles DR, Needleman I. A systematic review of definitions of periodontitis and methods that have been used to identify this disease. *Journal of clinical periodontology* 2009;**36**:458-467.

Figures Legends:

Figure 1: Prisma Flow Chart
Flow chart of the study selection process. A systematic review yielded 9760 reports. After removal of duplicates and the application of inclusion and exclusion criteria, 46 studies were included in the meta-analysis. Pubmed, Embase, Cochrane, Lilacs; Web of Science and manual search search strategies are illustrated in Supplementary material online, Appendices 1-2.

Figure 2: Association between Periodontitis (moderate to severe combined diagnosis) and Hypertension (Cross-sectional and Case-control studies)
Summary forrest plot for odds ratio of hypertension in relation to periodontitis diagnosis in cross-sectional and case-control studies (moderate to severe combined diagnosis). The random effects model was used and the relative size of the data markers indcates the weight of the sample size from each study. SE, standard error; IV, inverse variance; CI – confidence intervals.

Figure 3 A-C: Association between Periodontitis (Severe, Confident and non-Confident diagnosis) and Hypertension
Subgroup analysis forrest plots for odds ratio of hypertension in relation to periodontitis status in cross-sectional and case-control studies.Panel A: Severe periodontitis only group adjusted; Panel B: Analysis adjusted for confident definition of periodontitis as described in methods; Penel C: Analysis adjusted for non-confident definition of periodontitis as described in methods. The random effects model was used and the relative size of the data markers indcates the weight of the sample size from each study. SE, standard error; IV, inverse variance; CI – confidence intervals.

Figure 4: Periodontitis effect on Systolic Blood Pressure (SBP)
Summary forrest plot for change in systolic blood pressure (SBP) in relation to periodontitis status in cross-sectional and case-control studies. The random effects model was used, Weight Mean Difference (WMD) reported and the relative size of the data markers indcates the weight of the sample size from each study. SE, standard error; IV, inverse variance; CI – confidence intervals.

Figure 5: Periodontitis effect on Diastolic Blood Pressure (DBP)
Summary forrest plot for change in diastolic blood pressure (SBP) in relation to periodontitis status in cross-sectional and case-control studies. The random effects model was used, Weight Mean Difference (WMD) reported and the relative size of the data markers indcates the weight of the sample size from each study. SE, standard error; IV, inverse variance; CI – confidence intervals.

Figure 6: Some pathways through which Periodontitis could lead to Hypertension and Vascular Dysfunction.
Potential mechanisms causally linking periodontitis with hypertension and vascular dysfunction. NO- nitric oxide; O2-. S- superoxide; Th – T helper cells; ROS- reactive oxygen species

3.2. MATERIAL & METHODS AND RESULTS FROM 2ND ORIGINAL PUBLICATION

Reference: Muñoz Aguilera E, Leira Y, Miró Catalina Q, Orlandi M, Czesnikiewicz-Guzik M, Guzik TJ, et al. Is systemic inflammation a missing link between periodontitis and hypertension? Results from two large population-based surveys. Journal of Internal Medicine. 2021;289(4):532-46.

Title: Is systemic inflammation a missing link between periodontitis and hypertension? results from two large population-based surveys.

Authors: Muñoz Aguilera, E., Leira, Y., Miró Catalina, Q., Orlandi, M., Czesnikiewicz-Guzik, M., Guzik, T.J., Hingorani, A.D., Nart, J. and D'Aiuto, F.

Journal: Journal of Internal Medicine
Impact factor: 6.871
Journal quartile: Q1

R2: Is Systemic Inflammation a Missing Link Between Periodontitis and Hypertension? Results from Two Large Populations-based Surveys.

Running headline: Periodontitis, hypertension and systemic inflammation.

Authors: Eva Muñoz Aguilera, BDS, MClinDent*[1,2], Yago Leira, BDS, PhD*[1,3], Queralt Miró Catalina,BS, MS[4], Marco Orlandi, BDS, PhD[1], Marta Czesnikiewicz-Guzik, BDS, PhD[5,6], Tomasz J. Guzik, MD, PhD[7,8], Aaron D. Hingorani, MD, PhD[9], José Nart, BDS, PhD[2], Francesco D'Aiuto, DMD, PhD[§1]

*Equally first author
[§]Corresponding author

Affiliations:
[1]Periodontology Unit, UCL Eastman Dental Institute and Hospital, University College London, London, UK.
[2]Department of Periodontology, Universitat Internacional de Catalunya, Barcelona, Spain.
[3]Periodontology Unit, Faculty of Odontology, University of Santiago de Compostela & Medical-Surgical Dentistry (OMEQUI) Research Group, Health Research Institute of Santiago de Compostela (IDIS), Santiago de Compostela, Spain.
[4]Department of Statistics, Universitat Internacional de Catalunya, Barcelona, Spain
[5]Department of Periodontology and Oral Sciences Research Group, University of Glasgow Dental School, Glasgow, UK.
[6]Department of Experimental Dentistry and Dental Prophylaxis, Jagiellonian University, Krakow, Poland.
[7]Institute of Cardiovascular and Medical Sciences, University of Glasgow, Glasgow, UK.
[8]Department of Internal and Agricultural Medicine, Jagiellonian University, Krakow, Poland.
[9]Genetic Epidemiology, Institute of Cardiovascular Science, Faculty of Population Health Sciences, University College London, London, UK.

Corresponding author:
Prof. Francesco D'Aiuto
Periodontology Department, UCL Eastman Dental Institute
21 University Street, London, WC1E 6DE
Email: f.daiuto@ucl.ac.uk

Proofreader:
Dr Eva Muñoz Aguilera
Email: eva.aguilera.15@ucl.ac.uk

Abstract:

Objective: The primary objective was to investigate the relationship between periodontitis and hypertension in two independent large surveys. The secondary objective was to ascertain whether systemic inflammation had a mediation effect in the association.

Methods: This cross-sectional study analyzed representative samples of the US (n=3460; NHANES 2009/10) and Korean (n=4539; 2015 KNHANES VI-3) populations. The association between periodontitis (exposure), hypertension (outcome) and inflammatory markers [C-reactive protein (CRP) and white blood cell counts (WBC)] (mediators) was assessed using multivariate linear and logistic regression models and mediation analysis.

Results: Participants with periodontitis were more likely to have hypertension (NHANES:OR=1.3, 95%CI:1.0-1.6, p=0.025; KNHANES:OR=1.2, 95%CI:1.0-1.4, p=0.041) and actual systolic blood pressure ≥140 mm Hg (NHANES:OR=1.6, 95%CI:1.1-2.3, p<0.001; KNHANES:OR=1.3, 95%CI:1.0-1.6, p<0.031) than those without the disease. These associations were independent of age, gender, BMI, education level, smoking, alcohol consumption, creatinine, physical activity, presence of other comorbidities and confirmed in participants not taking antihypertensive medications. Diagnosis of periodontitis was directly associated with WBC (in both surveys- NHANES:β±SE=0.3±0.1, p<0.004; KNHANES:β±SE=0.3±0.1, p<0.001) and with CRP levels (in one survey: NHANES:β±SE=0.1±0.03, p<0.007; KNHANES:β±SE=0.1±0.04,p>0.213). Mediation analyses confirmed that CRP acted as a mediator in the association between periodontitis and hypertension in both populations (Mediated effect: NHANES:β±SE=0.010±0.003, p<0.001; KNHANES:β±SE=0.003±0.001, p=0.015). WBC acted as a mediator in the KNHANES (Mediated effect:β±SE=0.004±0.001, p=0.004) whilst in the NHANES its effect was dependent of CRP inclusion in the model (Mediated effect WBC+CRP:β±SE=0.002±0.001, p=0.001).

Conclusions: These findings suggest that periodontitis is closely linked to hypertension and systemic inflammation is, in part, a mediator of this association.

Key words: Periodontitis, Hypertension, High blood pressure, Systemic inflammation, CRP, Leukocytes

Introduction

Hypertension is a complex multifactorial disorder. Its prevalence exceeds 31% worldwide with more than 1.13 billion people affected (1). Elevated blood pressure (BP) is strongly linked to cardiovascular complications, increasing morbidity and mortality (2). Experimental and observational evidence supports a prominent role of systemic inflammation both in the initiation and progression of hypertension (3). The management of this condition, however, is still a challenge and it represents an increasing burden for society.

Periodontitis is one of the most common inflammatory disorders worldwide with >46% of adults in the US diagnosed with the disease (4). Strong evidence supports the role of a dysbiotic dental biofilm in the development of periodontitis. Further a cluster of modifiable risk factors are shared between periodontitis and leading non communicable diseases (NCDs) (cardiovascular diseases, cancer, chronic respiratory diseases and diabetes) (5).

Patients with periodontitis exhibit not only gingival inflammation but also endothelial dysfunction, increased bacterial burden (endotoxins and exotoxins dissemination), metabolic dysregulation, and systemic inflammation (6-9). A bi-directional link has been proposed between periodontitis and other metabolic disorders such as metabolic syndrome and diabetes (10, 11).

Hypertension has been linked to periodontitis but evidence from intervention trials is limited (12). A possible causal relationship between these two conditions has been proposed recently using Mendelian Randomization. This analysis confirmed a link between genetic variants linked to periodontitis and elevated BP phenotypes in a large UK population study (13). The exact mechanisms mediating this association remain unknown raising the question of whether inflammation or bacterial burden could play a prominent role. Given that systemic inflammatory biomarkers such as C-reactive protein (CRP) and leukocyte counts have been correlated with both periodontitis and hypertension, we hypothesized that systemic inflammation could be a mediator between the two diseases. Therefore, confirmation of this association in large independent studies with a focus on mediators need to be unraveled prior to undertaking interventional trials investigating the treatment of periodontitis as a target non-pharmacological treatment for hypertension. Accordingly, the primary aim of this study was to investigate the association between periodontitis and hypertension using two representative surveys of the US and Korean populations. The secondary aim was to ascertain the role of systemic inflammation in mediating this association.

Material and methods:

Two population-based surveys were analyzed and hereby reported in compliance with the Strengthening the Reporting of Observational studies in Epidemiology (STROBE) guidelines (Supplemental checklist).

Survey designs and study populations

Databases obtained from the US [2009/2010 National Health And Nutrition Examination Survey (NHANES)] and Korean [2015 VI-3 Korean National Health And Nutrition Examination Survey (KNHANES)] open repositories shared similar study design (stratified, multistage cluster probability sampling survey) (14, 15) and they were conducted by the statistical division of the National Centers for Disease Control and Prevention in US and Korea respectively. The study was conducted in accordance with the 1975 Declaration of Helsinki and participants provided written consent. These survey waves were selected for this study as were the first ones containing a detailed periodontal examination, measurements of average BP, serum concentrations of high sensitivity CRP (hs-CRP), and white blood cell counts (WBC).

Exclusion criteria used in the final sample analysis were (i) age <30 years in the NHANES (N=6,451) as no periodontal data was collected and age <19 years in the KNHANES (N=1,345); (ii) pregnancy (NHANES, N=23; KNHANES, N=29); (iii) lack of data on hs-CRP (NHANES, N=137; KNHANES, N=243); (iv) lack of data on BP (NHANES, N=123; KNHANES, N=29) and (v) lack of periodontal data for any other reasons (NHANES, N=343, KNHANES, N=792). From a total of 10,537 participants in the NHANES 2009/2010 and 6,977 in the KNHANES VI-3, the final samples included in this analysis were of 3,460 and 4,539 participants respectively. These populations refer to a representative sample of just over 128 millions of US and 33 millions of Korean citizens.

We extracted data on sociodemographic, health lifestyle behavioral factors, anthropometric measurements, medical history, oral examination, mean BP and biochemical parameters (eTable 1).

Blood pressure measurements

In both cross-sectional studies, sitting BP was measured using a standardized protocol (16). Average measurements of systolic and diastolic arterial pressure (SBP and DBP) were obtained from three consecutive readings. Participants were then categorized as normal, prehypertensive, and hypertensive according to the Joint National Committee 7 guidelines (17). Further, hypertension was

defined as values of SBP ≥140 mmHg or DBP ≥90 mmHg or the use of antihypertensive medication (18). The number of participants taking antihypertensive medications was also calculated.

Periodontal examination and dental exposure variables

The analysis was conducted using both established case definitions as well as continuous measures (full mouth indices) of periodontitis. However, different protocols were used. In the NHANES, a full-mouth periodontal assessment was carried out at six sites per tooth and periodontitis (exposure) was defined as mild, moderate or severe (19). Continuous aggregate dental variables (number and percentage of sites) were then created to indicate a) the extent of periodontal lesions with probing pocket depths (PPD) of ≥ 4mm, ≥5mm, ≥6 mm and b) the extent of loss of periodontal tissue attachment (AL) of ≥3mm, ≥ 4 mm, ≥5mm, ≥6mm as previously described (20).

In the KNHANES study, participants presenting with CPI scores of 3 and 4 (at least in one sextant) were defined as having worse periodontal status, whereas those presenting scores of 0, 1 and 2 represented controls with better periodontal status. Continuous measures of periodontal lesions were then created as follows: a) CPI cumulative score (the sum of only CPI scores of 3 or 4 of all sextants); and b) CPI continuous score (sum of all CPI scores of all sextants) as previously described (21).

Laboratory analysis

Biochemical parameters were retrieved from both surveys including plasma glucose (mg/dL), insulin levels (uIU/mL), glycated hemoglobin [HbA1c (%)], total cholesterol (mg/dL), high- and low-density lipoprotein cholesterol levels [HDL and LDL (mg/dL)], triglyceride levels (mg/dL), creatinine (mg/dL), hs-CRP (mg/dL), and WBCs (thous/μL)].

Statistical analysis

Data analyses were performed with STATA version 15.0 (StataCorp, College Station, Tex, US) and R Software (version 3.5.2). Continuous variables are reported as mean ± standard error (SE), whereas categorical variables are expressed as percentages. Simple differences between participants with or without periodontitis were assessed by independent t-test (for continuous variables) or χ square test (for categorical variables). Normality assumptions were checked, and a logarithmic transformation of hs-CRP was used for parametric analyses. Different measures of oral disease exposure (categorical and continuous) were adopted to test the association between periodontal status and BP. Further, circulating levels of WBC and hs-CRP were used as biomarkers of systemic inflammation and possible mediators of the association between periodontal status and hypertension. Univariate

analyses were performed for all continuous variables comparing the groups of participants with periodontitis/worse periodontal status and the rest of the study sample. All those variables with statistically significant associations were then used in the multivariate models.

Multivariate logistic regression models were created to test potential associations between periodontitis case definitions or continuous measures of periodontal lesions with hypertension, SBP ≥140 mmHg or hs-CRP>2mg/L as outcome variables. Multivariate linear regression models were then constructed to investigate the association between periodontal (both categorical and continuous) and arterial BP (mean SBP/DBP) variables. Similar analyses were performed with hs-CRP and WBC values (exposures) and hypertension (as categorical or continuous outcomes). Odds ratios (ORs) and 95% confidence intervals (CI) were calculated as well as β coefficient with standard errors. A fully adjusted model (Model 1) included age, gender, ethnicity, smoking, education level, and chronic medical conditions as covariates (as previously reported) in both surveys (18). In addition to these, body mass index (BMI), alcohol consumption, creatinine and physical activity were included in the multivariable models of the KNHANES survey (as they all presented univariate association with the outcome variables).

Sensitivity analyses in the subgroup of participants not taking antihypertensive medications were also performed (Model 2) (NHANES, N=2486; KNHANES, N=3270).

Structural equation modelling (SEM) was then used to estimate whether the association between periodontitis and hypertension was mediated by WBC or CRP using R Software (22). Four different and prespecified routes were used, direct (route 1) and indirect (route 2, 3, 4) mediation effects with their 95% CI were estimated:

Route 1: Periodontitis (exposure) → Hypertension (outcome).

Route 2: Periodontitis (exposure) → WBC (mediator) → Hypertension (outcome).

Route 3: Periodontitis (exposure) → Log CRP (mediator) → Hypertension (outcome).

Route 4: Periodontitis (exposure) → WBC (mediator) → Log CRP (mediator) → Hypertension (outcome).

Results

Characteristics of study populations

Participants with periodontitis were predominantly men (NHANES, 60%; KNHANES, 57%), older than 50 years of age, increased number of current smokers (NHANES, 52% vs 36%; KNHANES, 23% vs 15%), of lower education background and higher prevalence of diabetes (NHANES, 12% vs 6%; KNHANES, 10% vs 5%) than participants without periodontitis (Table 1). Almost a doubled prevalence of hypertension (NHANES, 42% vs 25%; KNHANES, 39% vs 19%) and anti-hypertensive medication (NHANES, 31% vs 19%; KNHANES, 25% vs 12%) were observed in patients with periodontitis. Similarly, participants with periodontitis had higher values of SBP (6.4 mm Hg higher in NHANES and 7.3 mm Hg higher in KNHANES) than survey participants without periodontitis. In the NHANES survey, Mexican and non-Hispanic black presented with the greatest prevalence of periodontitis. Lastly, when other traditional cardiovascular risk factors were assessed, patients with periodontitis exhibited greater values of glucose, triglycerides, hs-CRP and WBC in both surveys when compared to those without periodontitis, with BMI being higher in periodontitis patients only in KNHANES (all $p<0.001$).

Logistic Regression Analyses

Multiple logistic regression models confirmed that among participants with periodontitis and worse periodontal status, the adjusted odds of hypertension were 1.3 (95%CI 1.0-1.6) in the NHANES and 1.2 (95%CI 1.0-1.4) in the KNHANES populations, respectively (Table 2). Greater odds of hypertension in patients with periodontitis and worse periodontal status (CPI 3-4) were observed in the subgroup of participants not taking anti-hypertensive medications (NHANES: OR=1.4, 95%CI 1.0-1.8, N=2486; KNHANES: OR=1.3, 95%CI 0.9-1.7, N=3270). Similar associations were found between diagnosis of periodontitis and worse periodontal status (CPI 3-4) and SBP≥140 values in both populations (NHANES: OR=1.6, 95%CI 1.2-2.1; KNHANES: OR=1.3, 95%CI 1.0-1.6) with greater odds in participants with more severe periodontitis. These findings were consistent in those participants not taking anti-hypertensive medications (Model 2) although the estimates were smaller than those observed in the whole sample (Table 2). Indeed, NHANES participants with severe periodontitis also presented with more than twice increased likelihood of SBP≥140 mm Hg in model 1 (OR=2.5 95%CI 1.7-3.6) and model 2 (OR=2.3 95%CI 1.4-3.6), respectively (Table 2).

When hs-CRP or WBC were introduced as independent exposure variables, the odds of hypertension and SBP≥140 mm Hg ranged from 1.0 to 1.4 in the US and from 1.0 to 1.1 in the Korean survey. Only

some continuous measurements of periodontitis (mean PPD and mean CAL) were associated with greater odds of hypertension, SBP≥140 mm Hg, and of hs-CRP≥2 mg/l in the fully adjusted model and in those participants not taking antihypertensive medications (Figure 1).

Linear Regression Analyses

Linear regression analyses confirmed that periodontitis (assessed both as categorical and continuous variables) was associated with mean SBP. These findings were confirmed in the subgroup of participants not taking antihypertensive medications in the US survey (Table 3). In the Korean survey, the cumulative CPI score was consistently associated with SBP and DBP and this was also confirmed in participants not taking BP medications. Higher WBC counts were associated with mean SBP in both surveys, whilst higher hs-CRP levels were associated with SBP and DBP only in the US study (Table 3). Further, we observed a negative association between DBP and the number or percentage of gingival sites with attachment loss of ≥ 3 mm and of sites with probing depth ≥ 6 mm in the NHANES (model 2) and with the cumulative CPI score in the KNANES (models 1 and 2) (Table 3). Lastly, both US and Korean participants with severe periodontitis or worse periodontal status (CPI 3-4) exhibited greater systemic inflammation as assessed by hs-CRP serum levels and by WBC when compared to those without periodontitis or better periodontal status (CPI 0-2) and this difference was independent of other common confounders (Table 4).

Mediation Analyses

The association between periodontitis and hypertension (categorical) was mediated by CRP (β±SE=0.010±0.003; p<0.001) in the NHANES dataset, whilst WBC (β ±SE=0.001±0.001; p=0.221) was only an indirect mediator of the association (indirect route linked to hs-CRP) (Model A, unadjusted) (Figure 2 A and eTable 2). When repeating the same analysis in the KNHANES database, both hs-CRP (β±SE=0.003±0.001; p=0.015) and WBC (β±SE=0.004±0.001; p=0.004) acted as mediators of the association between worse periodontal status and hypertension (Figure 2 B and eTable 2). Models B, D, C replicated these results when the analyses applied for continuous periodontal (PPD, CAL in NHANES, and CPI continuous in KNANES) and BP (SBP) variables, in both adjusted and unadjusted models (eTable 2).

Discussion:

The analysis of two of the largest population-surveys with available dental and general health data demonstrated that both categorical and continuous measures of periodontitis were consistently associated with hypertension and SBP independent of other common cardiovascular risk factors. Participants in the US with severe periodontitis had higher odds for SBP≥140 mm Hg when compared to participants without periodontitis and all findings were confirmed in participants not taking anti-hypertensive medications. Systemic inflammation defined by two commonly measured biomarkers (hs-CRP and WBC) was not only associated independently with periodontitis, SBP and diagnosis of hypertension but acted as a modest mediator of these associations.

This analysis confirmed that participants with periodontitis have a 20-60% greater chance of presenting also a concomitant diagnosis of hypertension and a 10% to 2.5 times greater chance of SBP≥140 mm Hg. This is consistent with previous studies reporting that patients with periodontitis have on average 4.5 higher mean SBP (95% CI: 2.88–6.11) than participants without periodontitis (12). In the present study, a higher mean SBP of 6.4 mm Hg (NHANES, 95%CI 5.3-7.4) and of 7.2 mm Hg (KNHANES, 95%CI 6.1-8.4) were observed when participants with periodontitis were compared to those without the disease. The magnitude of this association could have important public health implications if we consider that high sodium intake with the diet is linked to a 6.0mmHg higher average SBP (23). Further SBP is a strong independent risk predictor for coronary heart disease events, stroke, heart failure and end-stage renal disease (24, 25).

Negligible associations of measures of periodontitis with DBP have been reported (20, 26). Interestingly, we observed a negative linear association between two measures of continuous disease and DBP in both datasets. This observation has not been reported previously, and it seems to be in contrast with the findings related to SBP. At this stage it is speculative to suggest a biological explanation of these findings. Authors consider important the role of residual confounding from other traditional risk factors variables (i.e. age, gender, ethnicity) as the estimates of association between DBP and some of the continuous measures of periodontitis tended to be greater in multivariate fully adjusted models. DBP is not considered on its own as a strong predictor for CVD events (27, 28), and the role of inflammation on affecting this measure of blood pressure is unclear. Further research should be conducted to ascertain the degree of association between diastolic pressure and periodontal inflammation as well as investigate potential biological mechanisms linking them.

When comparing the findings between the two surveys, a stronger association of categorical and continuous variables of periodontitis and gingival inflammation with hypertension and SBP were observed in the US dataset when compared with the Korean data survey. Similar findings were found for the association between biomarkers of inflammation (CRP and leucocytes counts) with measures of arterial blood pressure. Whilst the two sample populations present ethnic and socio-economic differences including a higher proportion of current smokers, adiposity and chronic medical conditions in the US population, authors believe that the different clinical measures of periodontitis recorded in the surveys could influence the results of the analyses.

A recent intervention study assessing the impact of periodontitis treatment on arterial blood pressure confirmed a substantial reduction of SBP after 2 months (mean difference of 11.1 mmHg) (13). This preliminary evidence suggests that periodontal treatment could represent a novel non-pharmacological intervention for hypertension of similar magnitude of other lifestyles adjustments (weight loss, increasing physical activity, salt or alcohol intake reduction or smoking cessation) with an average reduction of SBP ranging from 4.6 to 6.4 mmHg (29-31). However, larger and longer RCTs are needed.

Several lines of evidence now implicate inflammation in the development and progression of vascular diseases. For the last 3 decades inflammation has been recognized as a common denominator of early vascular dysfunction, leading onto the development of atheroma and vascular complications (32). Recent proof of concept evidence suggests that targeting upstream inflammation by selective drugs results in reduced morbidity and mortality (33). This could also be applicable in hypertension. Experimental and human studies have documented several pathways by which elevated inflammatory markers such as CRP and circulating leukocytes are associated with an increased risk of incident hypertension including a derangement of the renin-angiotensin system, increased oxidative stress, and downregulation of nitric oxide leading to increased endothelial stiffness and dysfunction (34). A recent review identified a number of potential sources of extravascular inflammation including periodontitis as a potential factor influencing vascular risk (32). It is now well documented that patients with periodontitis have elevated levels of CRP and WBC (7, 35).

In the mediation analysis, our findings suggest that CRP and WBC mediate partly the association between periodontitis and hypertension, although the effect is rather modest in nature (only 2% of the association explained by the model for the Korean survey whilst up to 7% in the US survey). Similar

findings were recently reported for CRP (5.4%), WBC (4.2%) and ferritin (10.2%) as mediators of the total association between a continuous measure of periodontitis and high/uncontrolled BP (≥130/80 mmHg) (36). An alternative pathway implicated in hypertension and cardiovascular injury relates to the activation of innate and adaptive immune cells such as monocyte/macrophages, and B and T lymphocytes (37). Damage-activated molecular patterns from the vasculature and Pathogens-activated molecular patterns from opportunistic diseases such as periodontitis can exacerbate the inflammatory cascade by activation of Th1 and Th17 lymphocytes, with kidneys and vasculature injuries aggravating a pro-hypertensive status, which results in progressive raised BP (38, 39).

Our analyses point perhaps towards a more prominent role of the gut and oral microbiome and their dysbiosis on hypertension (40). Periodontal pathogens may well play a role influencing the gut microbiome as well as exerting a direct vascular effect. Swallowing gram-negative oral bacteria or their end-products may trigger metabolic endotoxemia and systemic inflammation contributing to cardio-metabolic disorders (41). Lastly, experimental studies confirmed that periodontal bacteria can cause lower nitric oxide bioavailability and vascular dysfunction (42) and patients with periodontitis exhibit less nitrate-reducing bacteria (43). These novel mechanistic hypotheses warrant further investigation.

Cross-sectional designs preclude any inference on a possible temporal and/or causal association between periodontitis and hypertension. In the attempt of mitigating this limitation, we performed the analysis in two large surveys as to identify common patterns of association and minimizing spurious findings. Two different periodontal assessments and case definitions were adopted in each survey which could be considered a limitation but could also show that the association remains significant irrespectively. While in the NHANES, a recognized case definition was used (19), in the Korean survey a simplified clinical index (CPI) was selected, which is known to have risks of overestimation of the extent but underestimation of the prevalence of periodontitis (44). In the attempt to overcome some of these limitations we included a panel of measures of periodontal lesions to detect whether simple categorical associations were replicated when using other exposure variables. Another limitation to consider is the effect of antihypertensive medications on gingival inflammation and increased probing depths as well as on the overall association between periodontitis and hypertension (45). Sensitivity analyses were therefore performed by repeating the models in the group of participants not taking antihypertensive medications. We cannot however exclude that our

analyses missed some common risk determinants for hypertension (abdominal obesity, salt intake, use of anti-inflammatory drugs, hormone treatments, and stress) as well as unmeasured confounders associated with both periodontitis and hypertension (residual confounding). Future mechanistic and clinical studies should investigate further the role of periodontal-driven systemic inflammation and microbial burden as a risk factor for the development and management of hypertension and its complications.

Conclusion

Periodontitis is closely linked to hypertension and low-grade inflammation could be a key mediator in the association. Further interventional studies are needed to ascertain whether the treatment of periodontitis, leading to a decrease in systemic inflammation, may represent a novel non-pharmacological intervention in hypertension management.

Acknowledgments: The study was designed and carried out by E Muñoz Aguilera, Y Leira and F D'Aiuto. These three authors together with Q Miro performed the statistical analyses and interpreted the results. E Muñoz Aguilera, Y Leira and F D'Aiuto drafted the manuscript. J Nart, M Orlandi, M Czesnikiewicz-Guzik, TJ. Guzik, AD. Hingorani, provided critical interpretation and revision of the manuscript. All gave final approval and agreed to be accountable for all aspects of work ensuring integrity and accuracy.

Sources of Funding: We would like to acknowledge that contribution of this work was undertaken at UCLH/UCL who received a proportion of funding from the Department of Health's NIHR Biomedical Research Centre funding scheme. Dr. Leira holds a Senior Clinical Research Fellowship supported by the UCL Biomedical Research Centre who receives funding from the NIHR. TJG is funded by European Research Council (InflammaTENSION; ERC-CoG-726318) and ERA-NET CVD (PLAQUEFIGHT; 01KL1808 to T.J.G. / NCBiR, Poland)

Patient and Public Involvement:

This research was done without patient involvement. Patients were not invited to comment on the study design and were not consulted to develop patient relevant outcomes or interpret the results. Patients were not invited to contribute to the writing or editing of this document for readability or accuracy.

Disclosures: None

What is already known about this subject?	Consistent evidence suggests a direct relationship between periodontitis and hypertension. Poor oral health is linked to greater systemic inflammation and increased odds of hypertension. A linear association between systolic blood pressure and various oral health indices confirm these findings.
What does this study add?	Periodontitis increases the odds of hypertension by 20-60% in two large populations and systemic inflammation as assessed by peripheral levels of CRP and WBC acts as a biological mediator of this association.
How might this impact on clinical practice?	Oral health promotion could result in reduced systemic inflammation and it may represent a novel non-pharmacological intervention in hypertension management and its complications.

References

1. Zhou B, Bentham J, Di Cesare M, Bixby H, Danaei G, Cowan MJ, et al. Worldwide trends in blood pressure from 1975 to 2015: a pooled analysis of 1479 population-based measurement studies with 19· 1 million participants. The Lancet. 2017;389(10064):37-55.
2. Kirkland EB, Heincelman M, Bishu KG, Schumann SO, Schreiner A, Axon RN, et al. Trends in healthcare expenditures among US adults with hypertension: national estimates, 2003–2014. Journal of the American Heart Association. 2018;7(11):e008731.
3. Drummond GR, Vinh A, Guzik TJ, Sobey CG. Immune mechanisms of hypertension. Nature Reviews Immunology. 2019;19(8):517-32.
4. Eke PI, Dye BA, Wei L, Slade GD, Thornton-Evans GO, Borgnakke WS, et al. Update on prevalence of periodontitis in adults in the United States: NHANES 2009 to 2012. J Periodontol. 2015;86:611-22.
5. Sanz M, Marco del Castillo A, Jepsen S, Gonzalez-Juanatey JR, D'Aiuto F, Bouchard P, et al. Periodontitis and cardiovascular diseases: Consensus report. J Clin Periodontol. 2020;47(3):268-88.
6. Desvarieux M, Demmer RT, Jacobs Jr DR, Rundek T, Boden-Albala B, Sacco RL, et al. Periodontal bacteria and hypertension: the oral infections and vascular disease epidemiology study (INVEST). J Hypertens. 2010;28(7):1413.
7. Paraskevas S, Huizinga JD, Loos BG. A systematic review and meta-analyses on C-reactive protein in relation to periodontitis. J Clin Periodontol. 2008;35:277-90.
8. Orlandi M, Suvan J, Petrie A, Donos N, Masi S, Hingorani A, et al. Association between periodontal disease and its treatment, flow-mediated dilatation and carotid intima-media thickness: a systematic review and meta-analysis. Atherosclerosis. 2014;236(1):39-46.
9. Tonetti MS, Dyke TE. Periodontitis and atherosclerotic cardiovascular disease: consensus report of the Joint EFP/AAP Workshop on Periodontitis and Systemic Diseases. J Clin Periodontol. 2013;40.
10. Preshaw P, Alba A, Herrera D, Jepsen S, Konstantinidis A, Makrilakis K, et al. Periodontitis and diabetes: a two-way relationship. Diabetologia. 2012;55(1):21-31.
11. Jepsen S, Suvan J, Deschner J. The association of periodontal diseases with metabolic syndrome and obesity. Periodontology 2000. 2020;83(1):125-53.
12. Muñoz Aguilera E, Suvan J, Buti J, Czesnikiewicz-Guzik M, Barbosa Ribeiro A, Orlandi M, et al. Periodontitis is associated with hypertension: a systematic review and meta-analysis. Cardiovasc Res. 2020;116(1):28-39.
13. Czesnikiewicz-Guzik M, Osmenda G, Siedlinski M, Nosalski R, Pelka P, Nowakowski D, et al. Causal association between periodontitis and hypertension: evidence from Mendelian randomization and a randomized controlled trial of non-surgical periodontal therapy. Eur Heart J. 2019;40(42):3459-70.
14. Johnson CL, Paulose-Ram R, Ogden CL, Carroll MD, Kruszan-Moran D, Dohrmann SM, et al. National health and nutrition examination survey. Analytic guidelines, 1999-2010. 2013.
15. Kweon S, Kim Y, Jang M-j, Kim Y, Kim K, Choi S, et al. Data resource profile: the Korea national health and nutrition examination survey (KNHANES). Int J Epidemiol. 2014;43(1):69-77.
16. Ostchega Y, Prineas RJ, Paulose-Ram R, Grim CM, Willard G, Collins D. National Health and Nutrition Examination Survey 1999-2000: effect of observer training and

protocol standardization on reducing blood pressure measurement error. J Clin Epidemiol. 2003;56(8):768-74.
17. Chobanian AV, Bakris GL, Black HR, Cushman WC, Green LA, Izzo Jr JL, et al. The seventh report of the joint national committee on prevention, detection, evaluation, and treatment of high blood pressure: the JNC 7 report. JAMA. 2003;289:2560-71.
18. Mills KT, Bundy JD, Kelly TN, Reed JE, Kearney PM, Reynolds K, et al. Global disparities of hypertension prevalence and control: a systematic analysis of population-based studies from 90 countries. Circulation. 2016;134(6):441-50.
19. Eke PI, Page RC, Wei L, Thornton-Evans G, Genco RJ. Update of the case definitions for population-based surveillance of periodontitis. J Periodontol. 2012;83(12):1449-54.
20. Tsakos G, Sabbah W, Hingorani AD, Netuveli G, Donos N, Watt RG, et al. Is periodontal inflammation associated with raised blood pressure? Evidence from a National US survey. J Hypertens. 2010;28:2386-93.
21. Masi S, Gkranias N, Li K, Salpea KD, Parkar M, Orlandi M, et al. Association between short leukocyte telomere length, endotoxemia, and severe periodontitis in people with diabetes: a cross-sectional survey. Diabetes Care. 2014;37(4):1140-7.
22. Team RC. R: a language and environment for statistical computing. R Foundation for Statistical Computing, Vienna. http s. www R-proje ct org. 2017.
23. Elliott P, Stamler J, Nichols R, Dyer AR, Stamler R, Kesteloot H, et al. Intersalt revisited: further analyses of 24 hour sodium excretion and blood pressure within and across populations. BMJ. 1996;312(7041):1249-53.
24. Collins R, Peto R, MacMahon S, Godwin J, Qizilbash N, Hebert P, et al. Blood pressure, stroke, and coronary heart disease: part 2, short-term reductions in blood pressure: overview of randomised drug trials in their epidemiological context. The Lancet. 1990;335(8693):827-38.
25. Stamler J, Stamler R, Neaton JD. Blood pressure, systolic and diastolic, and cardiovascular risks: US population data. Arch Intern Med. 1993;153(5):598-615.
26. Pietropaoli D, Del Pinto R, Ferri C, Wright Jr JT, Giannoni M, Ortu E, et al. Poor oral health and blood pressure control among US hypertensive adults: results from the National Health and Nutrition Examination Survey 2009 to 2014. Hypertension. 2018;72(6):1365-73.
27. Kannel WB, Gordon T, Schwartz MJ. Systolic versus diastolic blood pressure and risk of coronary heart disease: the Framingham study. The American journal of cardiology. 1971;27(4):335-46.
28. Domanski M, Mitchell G, Pfeffer M, Neaton JD, Norman J, Svendsen K, et al. Pulse pressure and cardiovascular disease–related mortality: follow-up study of the Multiple Risk Factor Intervention Trial (MRFIT). JAMA. 2002;287(20):2677-83.
29. Sacks FM, Svetkey LP, Vollmer WM, Appel LJ, Bray GA, Harsha D, et al. Effects on blood pressure of reduced dietary sodium and the Dietary Approaches to Stop Hypertension (DASH) diet. N Engl J Med. 2001;344(1):3-10.
30. Neter JE, Stam BE, Kok FJ, Grobbee DE, Geleijnse JM. Influence of weight reduction on blood pressure: a meta-analysis of randomized controlled trials. Hypertension. 2003;42(5):878-84.
31. Whelton SP, Chin A, Xin X, He J. Effect of aerobic exercise on blood pressure: a meta-analysis of randomized, controlled trials. Ann Intern Med. 2002;136(7):493-503.
32. Libby P, Loscalzo J, Ridker PM, Farkouh ME, Hsue PY, Fuster V, et al. Inflammation, immunity, and infection in atherothrombosis: JACC review topic of the week. J Am Coll Cardiol. 2018;72(17):2071-81.

33. Ridker PM, MacFadyen JG, Everett BM, Libby P, Thuren T, Glynn RJ, et al. Relationship of C-reactive protein reduction to cardiovascular event reduction following treatment with canakinumab: a secondary analysis from the CANTOS randomised controlled trial. The Lancet. 2018;391(10118):319-28.
34. Guzik TJ, Touyz RM. Oxidative Stress, Inflammation, and Vascular Aging in Hypertension. Hypertension. 2017;70(4):660-7. doi: 10.1161/HYPERTENSIONAHA.117.07802. PubMed PMID: 28784646.
35. Loos BG. Systemic markers of inflammation in periodontitis. J Periodontol. 2005;76:2106-15.
36. Pietropaoli D, Del Pinto R, Ferri C, Marzo G, Giannoni M, Ortu E, et al. Association between periodontal inflammation and hypertension using periodontal inflamed surface area and bleeding on probing. J Clin Periodontol. 2020;47(2):160-72.
37. Guzik TJ, Hoch NE, Brown KA, McCann LA, Rahman A, Dikalov S, et al. Role of the T cell in the genesis of angiotensin II–induced hypertension and vascular dysfunction. J Exp Med. 2007;204(10):2449-60.
38. Krishnan SM, Ling YH, Huuskes BM, Ferens DM, Saini N, Chan CT, et al. Pharmacological inhibition of the NLRP3 inflammasome reduces blood pressure, renal damage, and dysfunction in salt-sensitive hypertension. Cardiovasc Res. 2019;115(4):776-87.
39. Wang H-X, Li W-J, Hou C-L, Lai S, Zhang Y-L, Tian C, et al. CD1d-dependent natural killer T cells attenuate angiotensin II-induced cardiac remodelling via IL-10 signalling in mice. Cardiovasc Res. 2019;115(1):83-93.
40. Santisteban MM, Qi Y, Zubcevic J, Kim S, Yang T, Shenoy V, et al. Hypertension-linked pathophysiological alterations in the gut. Circ Res. 2017;120(2):312-23.
41. Arimatsu K, Yamada H, Miyazawa H, Minagawa T, Nakajima M, Ryder MI, et al. Oral pathobiont induces systemic inflammation and metabolic changes associated with alteration of gut microbiota. Sci Rep. 2014;4:4828.
42. Velsko IM, Chukkapalli SS, Rivera MF, Lee J-Y, Chen H, Zheng D, et al. Active invasion of oral and aortic tissues by Porphyromonas gingivalis in mice causally links periodontitis and atherosclerosis. PLoS One. 2014;9(5).
43. Ashworth A, Cutler C, Farnham G, Liddle L, Burleigh M, Rodiles A, et al. Dietary intake of inorganic nitrate in vegetarians and omnivores and its impact on blood pressure, resting metabolic rate and the oral microbiome. Free Radic Biol Med. 2019;138:63-72.
44. Tran DT, Gay I, Du XL, Fu Y, Bebermeyer RD, Neumann AS, et al. Assessment of partial-mouth periodontal examination protocols for periodontitis surveillance. J Clin Periodontol. 2014;41(9):846-52.
45. Rodrigues M, Barbirato D, Luiz R, Scharfstein J, Salles G, Feres-Filho E. Effect of antihypertensive therapy with angiotensin-converting enzyme inhibitors on chronic periodontitis: a case–control study. Oral Dis. 2016;22(8):791-6.

3.3. MATERIAL & METHODS AND RESULTS FROM 3rd ORIGINAL PUBLICATION

Reference: Muñoz Aguilera E, Suvan J, Orlandi M, Miró Catalina Q, Nart J, D'Aiuto F. Association between periodontitis and blood pressure highlighted in systemically healthy individuals: Results from a nested case-control study. Hypertension. 2021;77(5):1765-74.

Title: Association between periodontitis and blood pressure highlighted in systemically healthy individuals. Results from a nested case-control study

Authors: Muñoz Aguilera, E., Suvan, J., Orlandi, M. Catalina, Nart, J., and D'Aiuto, F.

Journal: Hypertension
Impact factor: 7.713
Journal quartile: Q1

Association between periodontitis and blood pressure highlighted in systemically healthy individuals. Results from a nested case-control study.

Short title: Periodontitis and hypertension in systemically healthy individuals

Authors: Eva Munoz Aguilera[1,2], Jeanie Suvan[1], Marco Orlandi[1], Queralt Miró Catalina[2], Jose Nart[2], Francesco D'Aiuto[1]

Affiliations:

[1]Periodontology Unit, UCL Eastman Dental Institute and Hospital, University College London, London, UK

[2]Department of Periodontology, Universitat Internacional de Catalunya, Barcelona, Spain

Corresponding author:

Prof. Francesco D'Aiuto

Periodontology Department, UCL Eastman Dental Institute

21 University Street, London, WC1E 6DE

Email: f.daiuto@ucl.ac.uk

Phone: 00442034561108

Word count: 6000

Abstract

Recent evidence suggests hypertension and periodontitis are closely linked but limited data is available on the nature of the association. We aimed to investigate the relationship between periodontitis and mean arterial blood pressure in a sample of otherwise systemically healthy individuals. A case-control study including 250 cases (participants with periodontitis) and 250 controls (without periodontitis) was designed from a register of clinical trials conducted between 2000-2018 in a university setting. Cases were age, gender and body mass index balanced with controls. Linear, logistic regression and mediation models were planned to test the association between various periodontal measures and arterial blood pressure. We further investigated the role of systemic inflammation assessed by hsCRP and white cell counts. Cases presented with 3.36 mm Hg (95%CI, 0.91-5.82, p=0.007) higher mean systolic blood pressure (SBP) and 2.16 mm Hg (95%CI, 0.24-4.08, p=0.027) higher diastolic blood pressure (DBP) than controls. Diagnosis of periodontitis was associated with mean SBP (β=3.46±1.25, p=0.005) and greater odds of SBP≥140 mm Hg (OR=2.3, 95%CI:1.15-4.60, p=0.018) independent of common cardiovascular risk factors. Similar findings were observed when continuous measures of periodontal status were modelled against SBP. Measures of systemic inflammation although elevated in periodontitis were not found to be mediators of the association between periodontitis and arterial blood pressure values. Periodontitis is linked to higher systolic blood pressure in otherwise healthy individuals. Promotion of periodontal and systemic health strategies in the dental and medical setting could help reduce the burden of hypertension and its complications.

Key words: Periodontitis, bleeding on probing, mean blood pressure, systemic inflammation, case-control study, mediation analysis, hypertension.

1. Introduction

Elevated arterial blood pressure increases the risk of complications from cardiovascular diseases (CVD) such as stroke and myocardial infarction, with more than 7.6 million deaths accounted for every year and 143 million disability adjusted life-years.[1] It is estimated that more than 30% of the overall population suffers from hypertension, and this estimate increases with age.[2] A 15%–50% of individuals however are unaware they are affected by hypertension,[3] whilst many of those with a stablished diagnosis fail to achieve an optimal blood pressure control despite their prescribed medications.[2] The burden and cost of hypertension remain high for any given society. Inflammation is considered an important driver of vascular dysfunction and implicated in the development and progression of hypertension.[4,5]

Periodontitis is a common inflammatory disease caused by a dysbiotic biofilm and affecting the soft and hard tissues around teeth.[6] It is a chronic disease, usually spanning over decades of an individual's life and is characterised by gingival inflammation with associated alveolar bone loss which, if not arrested, will ultimately lead to tooth loss. Almost 750 million people (aged 15 to 99) worldwide present with moderate to severe symptoms of periodontitis,[7] plus the disease is linked to social inequality and it negatively affects patients' quality of life.[8]

Recent evidence suggests a possible causal link between periodontitis and hypertension.[9] Patients with periodontitis often present with higher arterial blood pressure values and a 30-70% higher chance to also present with hypertension,[10] especially when there is active gingival inflammation (i.e. with gingival bleeding).[11] Longitudinal and large interventional studies confirming the nature of this association and the exact pathogenetic mechanisms are scarce.

The aim of this study was to investigate the association between diagnosis of severe periodontitis and arterial blood pressure in a sample of otherwise healthy participants (without a confirmed diagnosis of hypertension). The primary objective was to assess office blood pressure values in patients with periodontitis (cases) compared to controls (participants without periodontitis) and whether a linear relationship exists between measures of periodontitis extent/severity with blood pressure values and whether basic measures of systemic inflammation mediate any association. Further, the prevalence of undiagnosed hypertension between cases and controls was explored.

2. Material and methods

The data that support the findings of this study are available from the corresponding author upon reasonable request.

2.1.Study design and population:

A nested case-control study was designed, including participants recruited at the UCL Eastman Dental Institute between the years 2001 – 2018. All participants provided written informed consent at the time of study participation including use of data for future analyses. Seventeen clinical trials with the same inclusion criteria (severe periodontitis) or control (no periodontitis) and full-mouth periodontal assessment and arterial blood pressure measurements (same recording protocol) were screened for inclusion in this analysis. This study was approved by the local UCL Research Ethic Committee (Project ID: 16989/001).

Sample inclusion criteria:

Cases were individuals ≥18 years old who had been diagnosed with generalised severe periodontitis (defined as ≥50% of the teeth with probing pocket depths (PPD) of ≥5mm and ≥30% marginal alveolar bone loss) and referred to the periodontal unit for management of their condition[12]. All participants were otherwise systemically healthy (no other systemic condition, as per medical history assessment/interview) and had not undertaken periodontal therapy within 6 months of the specific study assessment.

Controls were individuals ≥18 years old attending the same hospital without a diagnosis of periodontitis (recruited from other dental units but with no active dental infections) who were equally systemically healthy (no other systemic condition, as per medical history assessment/interview).

Sample exclusion criteria:

Possible participants were excluded from this study if presenting with any of the following: i) active infectious diseases such as hepatitis, HIV or tuberculosis, ii) any confirmed systemic

diseases including diabetes, kidney, liver, cardiovascular diseases, hypertension, cancer or on any chronic medication and iii) pregnant or breastfeeding, iv) taking non-steroidal anti-inflammatory drugs on a regular basis or taking antibiotics within 3 months of assessment.

2..2. Periodontal examination:

Periodontal assessment used a standardised protocol carried out by calibrated examiners as previously described.[12] Baseline data on full mouth periodontal assessment was retrieved for all participants. Case definition of periodontitis was confirmed against the latest validated classification.[6] The full-mouth dental plaque (FMPS) and gingival bleeding (FMBS) scores were recorded and the following thresholds for localised (FMBS 10%-29%) and generalized gingival bleeding (FMBS >30%) were adopted. Periodontitis case definition was of generalised severe/stage III/IV periodontitis.[6] Continuous measures of severity and extent of periodontitis were created as follows: a) the extent of periodontal pockets with probing pocket depth (PPD) of ≥ 4mm, ≥5mm, ≥6 mm (number and percentage of sites) and b) extent of loss of periodontal attachment levels (CAL) of ≥3mm, ≥ 4 mm, ≥5mm, ≥6mm (number and percentage of sites).

2.3. Blood pressure assessment:

Office blood pressure measurements were obtained following a standardised protocol using an Omron device M5-1 (HEM-757A-E) by a trained person and recorded in triplicate for each participant as previously described.[13] The patients were advised not to exercise, smoke or consume any caffeine during the 30 min prior to their appointment. Upon arrival, the measurements were recorded after the patients were seated for 5 min and relaxed, with the back resting on the chair and the arm on a desk at the level of the right atrium. Average of the systolic and diastolic arterial pressure (SBP and DBP) readings taken to the nearest value were obtained and used as continuous variables. Unconfirmed hypertension diagnosis was

evaluated applying diagnostic thresholds of the US (values of SBP ≥130 mmHg or DBP ≥80 mmHg) and European (values of SBP ≥140 mmHg or DBP ≥90 mmHg) guidelines.[14,15]

2.4. Additional variables:

Socio-demographics information (age, gender, ethnicity), health lifestyle behaviour [smoking, physical activity (frequency of weekly sessions of being active through walking, cycling, sports and recreation)], and family history of cardiovascular diseases (whether or not any family member had heart or vascular disease) was retrieved from the medical history questionnaires. Anthropometric measurements (body max index: BMI) were collected by trained staff using a standard protocol.[12] Fasting venous blood samples were collected and analysed for White Cell Counts (WBC), high sensitivity C-reactive protein (hsCRP), total cholesterol, low- and high-density lipoprotein, glucose and triglycerides using standard biochemistry procedures and as previously described.[12]

2.5. Statistical Analysis:

A sample size calculation confirmed that a minimum of 248 participants per group was required to detect a difference of 3.5 mm Hg in mean Systolic blood pressure (SBP) between cases and controls, with a standard deviation of 12 mm Hg, to achieve a power of at least 90% assuming an alpha of 0.05.

Cases and controls were balanced based on age ± 3years, gender and BMI (Figure 1).[16] All data was checked for errors, entered in a single dataset and analysed using SPSS (version 25), STATA (version 15) and R Software (version 3.5.2). Descriptive statistics (comparisons between continuous variables used independent t-test/Mann Witney and between categorical variables used Chi squared test) was used to compare general variables and arterial blood pressure levels between cases and controls. Multivariate linear regression analysis was performed to investigate potential associations between: i) periodontitis (categorical and continuous definitions) or systemic markers of inflammation (i.e. hsCRP and WBC) as

exposures and mean SBP and DBP as outcomes, ii) periodontitis (exposure) and systemic markers of inflammation (outcome). Age, BMI, gender, ethnicity, smoking, physical activity and family history of CVD were used as confounders in each model. Similarly, multivariate logistic regression analysis was carried out to test the odds of the following categorical outcome variables: i) SBP≥140, ii) SBP≥130, iii) DBP≥90, iv) DBP≥80 mm Hg and 'undiagnosed' hypertension defined as v) per European guidelines[14] (SBP/DBP≥140/90mmHg) and vi) as per US guidelines[15] (SBP/DBP≥138/80mmHg) in relation to the different exposure variables, i.e. periodontitis diagnosis (categorical yes/no), hsCRP and WBC. β coefficients and ORs with 95% confidence intervals were calculated in unadjusted (Model 1) and adjusted models including age, BMI, gender, ethnicity, smoking, physical activity and family history of CVD (Model 2). A p-value of ≤0.05 were considered statistically significant. Mediation analyses of systemic inflammatory markers were performed if a positive association between CRP or WBC variables with periodontitis and SBP/DBP were found in linear/logistic regression models. Two different and prespecified routes were used, direct (route 1) and indirect (route 2) mediation effects with their 95% CI were estimated: Route 1: Periodontitis (exposure)→ Hypertension (outcome). Route 2: Periodontitis (exposure)→ WBC or hsCRP (mediators)→ Hypertension (outcome).

2. Results

Sample median age [IQR] was 35 [12] years with no substantial differences observed between cases (35 [9] years) and controls (34 [14] years) (p=0.345). Similar gender (p=0.929) and BMI (p=0.209) distributions were confirmed between cases and controls. Higher number of current smokers, with predominant Afro-American/Afro-Caribbean participants, reporting less physical activity and a higher percentage of family history of CVD were observed in cases when compared to controls. Participants with periodontitis exhibited increased glucose, LDL, CRP and WBC levels but lower HDL values when compared with controls (all, p<0.01) (Table 1).

Higher mean differences in SBP (3.36mmHg, 95%CI, 0.91-5.82, p=0.007) and DBP (2.02 mmHg, 95%CI, 0.24-4.08, p<0.027) were observed in cases when compared to controls.

A 14% of cases presented with SBP≥140 mmHg vs a 7% of the controls (p=0.021). Similarly, more than 43% of the cases presented with DBP≥80 mmHg vs 34% of the controls (p=0.035). The percentages of cases with SBP≥130 mmHg or DBP≥90 mmHg was greater than for the controls but not statistically significant (Table 1). Overall a 15.6% of the whole study participants presented with values of SBP/DBP in the range of hypertension (European definition): 17.2% of the cases and 14% of the controls (p=0.324), and a 45.6% (American definition): almost 50% of cases and a 41.6% of controls (p=0.073) (Table 1).

Linear regression analysis confirmed an association between case definition of periodontitis (categorical) and higher mean SBP after adjusting for common risk factors (*ß*±SE=3.46±1.25, *p=0.005*, Model 2) and to DBP (*ß*±SE=2.16±0.98, p=0.027, Model 1) (Table 2). Greater severity of periodontitis as assessed by measure of CAL, PPD and FMBS were associated with higher mean SBP (Models 1 and 2) and with higher mean DBP (Model 1) (Table 2). A similar linear association between different thresholds of periodontal lesions

(PPD thresholds) and SBP (Models 1 and 2) were noted but for different threshold of CAL, this was only seen in Model 1 (Table 2). Periodontitis (categorical and continuous variables) was associated with higher CRP (Model 1) and WBC (Model 1 and 2) levels (Table 2). Similarly, periodontitis (categorical) was associated with higher odds of SBP≥140 mm Hg (OR=1.98, 95%CI, 1.10-3.65, *p*=0.023, Model 1; OR=2.31, 95%CI, 1.17-4.67, *p*=0.018 (Model 2) and DBP≥ 80 mm Hg (OR=1.47, 95% CI, 1.03-2.12, *p*=0.035, Model 1) (Table 3). No significant association was found with SBP≥130 mm Hg, DBP≥90-mm Hg or Hypertension definition according to European or American guidelines.

When CRP and WBC were modelled as exposure variables, no associations were observed with SBP or DBP (Tables 2-3). Lastly, an association between SBP and increasing FMBS irrespective of periodontal diagnosis was observed in the multivariate fitted model for SBP according to bleeding status (Table 4). Participants with generalised bleeding presented with a 5 mmHg greater SBP than those with healthy gums (95%CI, 8.22-1.91, *p*=0.002) (Table 4).

Mediation analyses confirmed that WBC did not act as a mediator of the association between periodontitis (categorical) and SBP (continuous) in either in the unadjusted (β±SE= -0.00±1.21; *p*=0.994) or the fully adjusted (β±SE= -0.03±0.21; *p*=0.900) models (Route 2) (Tables S1 and S2). Similar results were observed when the model was replicated for continuous periodontal (FMBS, CAL, PPD) and categorical BP (SBP≥140 mm Hg) variables.

5. Discussion

The results of this study showed that systemically healthy individuals with periodontitis (cases) presented with higher mean SBP and DBP than participants without periodontitis (controls). Cases presented with more than twice higher likelihood of SBP≥140 mm Hg and almost 50% higher odds of DBP≥80 mm Hg than the controls.

A recent systematic review confirmed a 4.5 mm Hg SBP- and 2.5 mm Hg DBP greater arterial blood pressure values in the general population (including participants with and without other co-morbidities).[10] In a previously reported age- and sex- matched case-control study looking at the association of periodontitis, systemic inflammation and endothelial function, greater differences in SBP (7 mm Hg) and CRP (1.3mg/L) levels between the cases and controls were observed.[17]

Elevated blood pressure remains the main risk factor for heart failure, atrial fibrillation, chronic kidney disease, heart valve diseases, aortic syndromes, and dementia, in addition to coronary heart disease and stroke.[18] It is now understood that biologically normal blood pressure levels are lower than what previously fell within a normal rangel.[15] The observed differences of SBP/DBP between cases and control in this study could be clinically relevant and might represented an overlooked mechanism linking periodontitis with increased future CVD risk.[19]

In agreement with previous studies,[20,21] this study showed that irrespective of periodontal status, bleeding gums was associated with SBP. Similarly, a more recent secondary analysis of NHANES III also reported a 2.6 mm Hg higher mean SBP for gingivitis, also independently associated with 40% greater odds of high/uncontrolled BP.[11] Bleeding gums, the earliest sign of periodontal diseases, has also been linked to increased systemic biomarkers and vascular changes.[22,23] Due to its easy detection by patients and clinicians could represent a valuable parameter in routine BP screening protocols.

Several mechanisms underlying the links between gingival diseases and hypertension have been proposed with dysbiotic subgingival microbiome triggering low-grade systemic inflammation and oxidative stress representing the main pathways.[24] Periodontitis patients express increased local and systemic inflammatory markers such as CRP, TNF- α, neutrophilic enzymes, WBC counts and disparity in T cell subtypes, but also neutrophil dysfunction, which are all mechanisms resulting in vascular changes and endothelial dysfunction.[19,25,26] The presence of periodontal pathogens have been linked to hypertension in epidemiological studies.[27] Pre-clinical evidence originated by experimental animal models, including immunisations with P. gingivalis lysate and Lipopolysaccharide (LPS) - endotoxin from other gram-negative bacteria caused prolonged T-cell activation and elicited increased levels of CRP, TNF-α, and IL-1β, resulting in increased blood pressure.[28] Interaction between oral-gut microbiome can also contribute to amplification of inflammation and metabolic changes.[29] Recent evidence implicates oral bacteria in the nitrate-nitrite-nitric oxide (NO) pathway and pathogenesis of hypertension,[30] with high concentrations of nitrite-reductase bacteria increasing systemic NO and having an effect of lowering SBP.[31]

In the current study, hsCRP/WBC as a proxy of systemic inflammation was associated with periodontitis but not with SBP/DBP. Additionally, WBC did not show a mediation effect between periodontitis and BP. These results are in partial disagreement with a recent analysis of cross-sectional data, based on national health surveys in US and Korea, where a 2-7% mediating effect of WBC and CRP was observed when examining the association between periodontitis and hypertension.[32] A possible explanation for these differences relates to an overall younger population of this study sample (35-years-old) vs 51- and 46- in the American and Korean populations, and possibly due to the systemically healthy status of this sample, when compared to representative samples of those populations, including systemic conditions. Nevertheless, an association of arterial BP with both continuous and categorical

measures of periodontitis in younger and systemically healthy individuals strengthens the evidence in favour of a causal association between the two diseases.[24]

A recent Mendelian Randomisation analysis and results from a short-term pilot RCT on periodontal treatment of resistant hypertensive patients corroborate these findings.[9] Single nucleotide polymorphisms (SNPs) in SIGLEC5, DEFA1A3, MTND1P5, and LOC107984137 loci GWAS-linked to periodontitis and BP phenotype were unravelled and a noticeable reduction in SBP/DBP, endothelial function as well as cytokines and activated T-cells subsets was observed 2-months following the treatment. Similarly, another RCT with 6-months follow-up on a pre-hypertensive population also observed a significant reduction in SBP/DBP following non-surgical periodontal treatment.[33] Oral health promotion strategies such as tooth brushing twice daily has demonstrated very effective, not only in managing and preventing most common oral conditions,[34] but in providing a powerful and affordable tool for hypertension control.[35] Notably, a 14% reduction in CV events have been observed with a 4.4 mm Hg reduction in SBP.[36] Preliminary evidence suggests that periodontal treatment in patients with type 2 diabetes, a common co-morbidity, could result in substantial long-term reduction of medical-related costs for healthcare systems.[37] Thus, given the importance of non-pharmacological and pharmacological blood pressure lowering strategies in decreasing CVD risk and mortality,[38] larger multicentre RCTs and health-economic analyses are warranted to further investigate the benefits of periodontal treatments on blood pressure prevention and control.

Elevated blood pressure is usually asymptomatic and best detected in screening programs or opportunistic measurements of BP, which confirm that a worryingly high number of individuals (> 50%) is unaware of a possible diagnosis of hypertension.[39] The presented study confirmed that a 15-45% of the sample could exhibit undetected hypertension (depending on whether a European or US guideline definition was used), with 54-55% of

these having periodontitis. In a recent cross-sectional study on the association of periodontitis and hypertension, a 15.9% of the study sample presented with undiagnosed high BP (based on a single office measurement), of which a 62.5% had periodontitis.[40] These data confirm that programmes of hypertension screening in the dental settings should not be underestimated.

Whilst this study improves the understanding around the association between periodontitis and arterial blood pressure, it is recognised some limitations exist. The study design and analysis may have introduced some bias namely through selection and assessment biases.[41] Further, in this study we did not account for other factors which might have impacted on blood pressure such as abdominal obesity, salt intake, use of anti-inflammatory drugs, hormone treatments or stress as well as additional oral diseases (i.e. caries). Future analyses should focus on existing or new epidemiological evidence (longitudinal studies) where all possible confounders are appropriately considered. Nevertheless, this study benefits from a robust research methodology in assessing the exposure (periodontitis) and outcome (blood pressure) and sufficient statistical power could have counteracted some of the limitations.[42] Further, using a balanced study design through matching for common confounders of arterial blood pressure facilitated analysis of comparable groups.[43]

6. Conclusions

This study expands current knowledge on the association between periodontitis and elevated blood pressure, pointing at the importance of this link in the generally healthy population. Oral health professionals could play a pivotal role in helping the medical community detecting and tackling the burden and consequences of hypertension.

7. Perspectives:

Periodontal treatments could be well-tolerated novelty non-pharmacological interventions for the management of hypertension. Particularly so when patients are informed that periodontal treatment could be beneficial not only for their oral health but also for their general health and wellbeing in return. Thus, future directions and broad implications of this work will involve liaison of dental and medical health professionals with the following objectives:

1. Raising awareness of the increased risk for high blood pressure among individuals with periodontal diseases.
2. Implementing hypertension screening systems by dental professionals and prompt referral to general practitioners.
3. Implementing periodontal diseases screening systems by medical professionals and referral to dental practitioners.
4. Providing advice for common risk factors: Healthy diet, smoking cessation, promoting physical activity, alcohol reduction, and diabetes management.
5. Early diagnosis and management of gingivitis and periodontitis. Effective prevention and treatment of these conditions is very cost-effective and has shown an effect in reduction of systemic markers of inflammation and improvement in endothelial function.
6. Future research will involve larger multicentre RCTs to test the effects on periodontal treatment on blood pressure levels.

8. Acknowledgments

We would like to acknowledge that contribution of this work received a proportion of funding from the UK Department of Health's National Institute for Health Research Biomedical Research Centre at University College London/University College London Hospitals.

9. Sources of funding

Dr. Orlandi holds a NIHR Clinical Lectureship. Prof. D'Aiuto held a Clinical Senior Lectureship Award supported by the UK Clinical Research Collaboration.

10. Conflict of interest/disclosures

Authors declare no conflicts of interest or disclosures.

11. References

1. Forouzanfar MH, Afshin A, Alexander LT, et al. Global, regional, and national comparative risk assessment of 79 behavioural, environmental and occupational, and metabolic risks or clusters of risks, 1990–2015: a systematic analysis for the Global Burden of Disease Study 2015. *The lancet.* 2016;388(10053):1659-1724.
2. Mills KT, Bundy JD, Kelly TN, et al. Global disparities of hypertension prevalence and control: a systematic analysis of population-based studies from 90 countries. *Circulation.* 2016;134(6):441-450.
3. Scholes S, Conolly A, Mindell JS. Income-based inequalities in hypertension and in undiagnosed hypertension: analysis of health survey for England data. *J Hypertens.* 2020;38(5):912-924.
4. Drummond GR, Vinh A, Guzik TJ, Sobey CG. Immune mechanisms of hypertension. *Nature Reviews Immunology.* 2019;19(8):517-532.
5. Jayedi A, Rahimi K, Bautista LE, Nazarzadeh M, Zargar MS, Shab-Bidar S. Inflammation markers and risk of developing hypertension: a meta-analysis of cohort studies. *Heart.* 2019;105(9):686-692.
6. Tonetti MS, Greenwell H, Kornman KS. Staging and grading of periodontitis: Framework and proposal of a new classification and case definition. *Journal of periodontology.* 2018;89:S159-S172.
7. Frencken JE, Sharma P, Stenhouse L, Green D, Laverty D, Dietrich T. Global epidemiology of dental caries and severe periodontitis–a comprehensive review. *J Clin Periodontol.* 2017;44(S18).
8. Ferreira M, Dias-Pereira A, Branco-de-Almeida L, Martins C, Paiva S. Impact of periodontal disease on quality of life: a systematic review. *J Periodontal Res.* 2017;52(4):651-665.
9. Czesnikiewicz-Guzik M, Osmenda G, Siedlinski M, et al. Causal association between periodontitis and hypertension: evidence from Mendelian randomization and a randomized controlled trial of non-surgical periodontal therapy. *Eur Heart J.* 2019;40(42):3459-3470.
10. Muñoz Aguilera E, Suvan J, Buti J, et al. Periodontitis is associated with hypertension: a systematic review and meta-analysis. *Cardiovasc Res.* 2020;116(1):28-39.
11. Pietropaoli D, Monaco A, D'Aiuto F, et al. Active gingival inflammation is linked to hypertension. *J Hypertens.* 2020;38(10):2018-2027.
12. D'Aiuto F, Parkar M, Nibali L, Suvan J, Lessem J, Tonetti MS. Periodontal infections cause changes in traditional and novel cardiovascular risk factors: results from a randomized controlled clinical trial. *Am Heart J.* 2006;151:977-984.
13. Muntner P, Shimbo D, Carey RM, et al. Measurement of blood pressure in humans: a scientific statement from the American Heart Association. *Hypertension.* 2019;73(5):e35-e66.
14. Williams B, Mancia G, Spiering W, et al. 2018 ESC/ESH Guidelines for the management of arterial hypertension: The Task Force for the management of arterial hypertension of the European Society of Cardiology (ESC) and the European Society of Hypertension (ESH). *European heart journal.* 2018;39(33):3021-3104.
15. Whelton PK, Carey RM, Aronow WS, et al. 2017 ACC/AHA/AAPA/ABC/ACPM/AGS/APhA/ASH/ASPC/NMA/PCNA guideline for the prevention, detection, evaluation, and management of high blood pressure in adults:

a report of the American College of Cardiology/American Heart Association Task Force on Clinical Practice Guidelines. *J Am Coll Cardiol.* 2018;71(19):e127-e248.
16. Grimes DA, Schulz KF. Bias and causal associations in observational research. *The lancet.* 2002;359(9302):248-252.
17. Amar S, Gokce N, Morgan S, Loukideli M, Van Dyke TE, Vita JA. Periodontal disease is associated with brachial artery endothelial dysfunction and systemic inflammation. *Arterioscler Thromb Vasc Biol.* 2003;23(7):1245-1249.
18. Fuchs FD, Whelton PK. High blood pressure and cardiovascular disease. *Hypertension.* 2020;75(2):285-292.
19. Sanz M, Marco del Castillo A, Jepsen S, et al. Periodontitis and cardiovascular diseases: Consensus report. *J Clin Periodontol.* 2020;47(3):268-288.
20. Pietropaoli D, Del Pinto R, Ferri C, et al. Association between periodontal inflammation and hypertension using periodontal inflamed surface area and bleeding on probing. *J Clin Periodontol.* 2020;47(2):160-172.
21. Tsakos G, Sabbah W, Hingorani AD, et al. Is periodontal inflammation associated with raised blood pressure? Evidence from a National US survey. *J Hypertens.* 2010;28:2386-2393.
22. Demmer RT, Papapanou PN, Jacobs Jr DR, Desvarieux M. Bleeding on probing differentially relates to bacterial profiles: the Oral Infections and Vascular Disease Epidemiology Study. *J Clin Periodontol.* 2008;35(6):479-486.
23. Bokhari SAH, Khan AA, Butt AK, et al. Periodontitis in coronary heart disease patients: strong association between bleeding on probing and systemic biomarkers. *Journal of clinical periodontology.* 2014;41(11):1048-1054.
24. Del Pinto R, Pietropaoli D, Munoz-Aguilera E, et al. Periodontitis and hypertension: is the association causal? *High Blood Press Cardiovasc Prev.* 2020;27:281-289.
25. Czesnikiewicz-Guzik M, Nosalski R, Mikolajczyk TP, et al. Th1-type immune responses to Porphyromonas gingivalis antigens exacerbate angiotensin II-dependent hypertension and vascular dysfunction. *British journal of pharmacology.* 2019;176(12):1922-1931.
26. Tonetti MS, D'aiuto F, Nibali L, et al. Treatment of periodontitis and endothelial function. *N Engl J Med.* 2007;356(9):911-920.
27. Desvarieux M, Demmer RT, Jacobs Jr DR, et al. Periodontal bacteria and hypertension: the oral infections and vascular disease epidemiology study (INVEST). *J Hypertens.* 2010;28(7):1413.
28. Libby P, Loscalzo J, Ridker PM, et al. Inflammation, immunity, and infection in atherothrombosis: JACC review topic of the week. *J Am Coll Cardiol.* 2018;72(17):2071-2081.
29. Arimatsu K, Yamada H, Miyazawa H, et al. Oral pathobiont induces systemic inflammation and metabolic changes associated with alteration of gut microbiota. *Sci Rep.* 2014;4:4828.
30. Goh CE, Trinh P, Colombo PC, et al. Association Between Nitrate-Reducing Oral Bacteria and Cardiometabolic Outcomes: Results From ORIGINS. *Journal of the American Heart Association.* 2019;8(23):e013324.
31. Tribble GD, Angelov N, Weltman R, et al. Frequency of tongue cleaning impacts the human tongue microbiome composition and enterosalivary circulation of nitrate. *Frontiers in cellular and infection microbiology.* 2019;9:39.

32. Muñoz Aguilera E, Leira Y, Miró Catalina Q, et al. Is systemic inflammation a missing link between periodontitis and hypertension? Results from two large population-based surveys. *Journal of Internal Medicine.*
33. Zhou Q-B, Xia W-H, Ren J, et al. Effect of Intensive Periodontal Therapy on Blood Pressure and Endothelial Microparticles in Patients With Prehypertension and Periodontitis: A Randomized Controlled Trial. *J Periodontol.* 2017;88(8):711-722.
34. Jepsen S, Blanco J, Buchalla W, et al. Prevention and control of dental caries and periodontal diseases at individual and population level: consensus report of group 3 of joint EFP/ORCA workshop on the boundaries between caries and periodontal diseases. *J Clin Periodontol.* 2017;44:S85-S93.
35. Choi HM, Han K, Park YG, Park JB. Associations among oral hygiene behavior and hypertension prevalence and control: the 2008 to 2010 Korea National Health and Nutrition Examination Survey. *Journal of periodontology.* 2015;86(7):866-873.
36. Law M, Morris J, Wald N. Use of blood pressure lowering drugs in the prevention of cardiovascular disease: meta-analysis of 147 randomised trials in the context of expectations from prospective epidemiological studies. *BMJ.* 2009;338:b1665.
37. Smits KP, Listl S, Plachokova AS, Van der Galien O, Kalmus O. Effect of periodontal treatment on diabetes-related healthcare costs: a retrospective study. *BMJ Open Diabetes Research and Care.* 2020;8(1):e001666.
38. Mahmood S, Shah KU, Khan TM, et al. Non-pharmacological management of hypertension: in the light of current research. *Irish Journal of Medical Science (1971-).* 2019;188(2):437-452.
39. Lindholt JS, Søgaard R. Population screening and intervention for vascular disease in Danish men (VIVA): a randomised controlled trial. *The Lancet.* 2017;390(10109):2256-2265.
40. Machado V, Aguilera EM, Botelho J, et al. Association between Periodontitis and High Blood Pressure: Results from the Study of Periodontal Health in Almada-Seixal (SoPHiAS). *Journal of Clinical Medicine.* 2020;9(5):1585.
41. Sedgwick P. Nested case-control studies: advantages and disadvantages. *Bmj.* 2014;348.
42. Gauderman WJ. Sample size requirements for matched case-control studies of gene–environment interaction. *Stat Med.* 2002;21(1):35-50.
43. Pearce N. Analysis of matched case-control studies. *BMJ.* 2016;352.

12. Novelty and significance

What is New?-

The link between periodontitis and high blood pressure was confirmed in systemically healthy individuals

What is Relevant?-

- Individuals with periodontitis and oherwise healthy presented with higher mean SBP/DBP and odds of SBP>140 mm Hg
- Bleeding gums was associated with higher mean SBP
- Undetected hypertension was a common finding among the population of study

Summary –

The risk of elevated blood pressure was highlighted in systmically healthy periodontal patients. Oral health professionals could play a crucial role in assisting in the screening and management of hypertension.

13. Figure legends:

Figure 1: Study flow chart diagram

Discussion

4. DISCUSSION

Summary of main findings

The three studies included in this doctoral thesis have produced new evidence contributing to a better understanding of the association between periodontitis and hypertension.

- *Diagnosis of Periodontitis is consistently associated with hypertension.*

The systematic review showed that diagnosis of moderate-severe periodontitis was associated with 22% increased odds of hypertension whereas severe periodontitis was associated with 49% increased odds, demonstrating a linear relationship between the two diseases. The magnitude of this association increased when a confident definition of periodontitis was used in the appraisal, and it resulted in greater odds (53%) of hypertension in patients with periodontitis compared to a 33% for a non-confident diagnosis. Meta-analysis of 3 cohort studies confirmed that diagnosis of periodontitis increased the likelihood of hypertension occurrence by 68%, although this was not statistically significant (P=0.14).

The findings from the systematic appraisal of the evidence were in line with the results observed then in the second and third study evaluation of this PhD. The cross-sectional data analysis of the Korean and American population confirmed that participants with periodontitis had 20-30% higher odds of hypertension than those without the disease, in addition to a 30-60% higher odds of SBP $\geq$ 140 mm Hg. These results were independent of age, gender, BMI, education level, smoking, alcohol consumption, creatinine, physical activity, presence of other comorbidities and confirmed in participants not taking antihypertensive medications differences. Similarly, the results from the nested case-control study, concerning a sample of otherwise systemically healthy individuals, showed that diagnosis of periodontitis was associated with 2.3 times higher likelihood of presenting with a SBP value equal or greater than 140 mm Hg independent of common cardiovascular risk factors.

- *There is a linear association between measures of periodontal health/disease and arterial blood pressure.*

In the cross-sectional study, the linear regression models carried out for both surveys confirmed that periodontitis was associated with mean SBP, and this was confirmed in the analyses for those participants without prescribed hypertensive drugs. Similarly, the linear regression models in the nested case-control study showed that periodontitis was associated with mean SBP. Interestingly, it was observed that the number and percentage of the measured sites with clinical attachment loss of ≥3 mm and of those with probing pocket depth ≥ 6 mm (in the American population) and the cumulative CPI score (in the Korean population) presented negatively associated to DBP.

The assessment of mean blood pressure levels according to the meta-analysis results showed that individuals with periodontitis exhibited greater mean SBP = 4.49 mm Hg; and DBP = 2.03 mm Hg when compared with non-periodontitis. The population of study in the different studies included in this meta-analysis comprised systemically healthy individuals as well as participants with other systemic conditions such as cardiovascular diseases and metabolic syndrome. When evaluating the mean blood pressure values for participants with and without periodontitis in the nested case-control study, we observed that the cases (periodontitis) presented with 3.36 mm Hg (P=0.007) higher mean SBP and 2.16 mm Hg (P=0.027) higher DBP than controls (non-periodontitis). Irrespective of the diagnosis of periodontitis, an association between SBP and increasing full mouth gingival bleeding score was observed in the multivariate fitted model. Participants with generalized bleeding presented with a 5 mm Hg (P=0.002) greater SBP than those with healthy gums.

- *Periodontitis triggers systemic inflammation and this could mediate its association with hypertension.*

The assessment of systemic markers of inflammation in the linear regression model carried out in the second study analysis revealed that diagnosis of periodontitis was directly associated with WBC in both surveys and with CRP levels only in the US

database. Additionally, mediation analysis showed that CRP mediated the association between periodontitis and hypertension in both populations. Nevertheless, WBC acted as a mediator in the Korean population though in the American population, WBC was not acting as an independent mediator, instead, its mediating effect depended on the inclusion of CRP in the same model. Similarly, the nested case-control study showed that periodontitis (categorical and continuous variables) was associated with higher WBC (adjusted and unadjusted models) and with CRP only in the unadjusted model. However, WBC did not act as a mediator of the association in this population.

- *There could be merit in suggesting opportunistic screening of hypertension in dental practice.*

The results of the case-control study revealed that a 15.6% of participants presented with values of SBP/DBP in the range of hypertension (European definition) without been aware of this possible risk. These percentages were slightly higher in those participants with a diagnosis of periodontitis (17.2% of the cases and 14% of the controls, *P*=0.324). When using the American definition, 45.6% of the whole study sample could have been at risk of having undiagnosed hypertension with almost 50% of cases and a 41.6% of controls presented with values of blood pressure higher than expected (*P*=0.073).

- *The current evidence suggests that treatment of periodontitis could result in reduction in blood pressure.*

After a comprehensive assessment of the interventional data based on 5 out of the 12 interventional studies, confirmed a decrease in BP levels following treatment of periodontitis. A reduction in SBP (ranging between 3- and 12.5 mm Hg) and from no changes to a 10 mm Hg reduction in DBP were reported in these studies suggesting a causal association between periodontitis and hypertension.

What does this PhD add to the current evidence and how does compare to it?

Collectively, the three study designs included in this thesis consistently demonstrated that periodontitis poses a higher risk for hypertension and higher systolic blood pressure, and that individuals with periodontitis have higher mean systolic and diastolic blood pressure values than those without periodontitis, irrespective of systemic health status. Additionally, systemic inflammatory markers such as CRP and WBC were elevated in periodontal patients and associated to periodontitis and hypertension case definitions. Moreover, systemic inflammation acted as a driver of this association in the cross-sectional study of the two large populations of USA and Korea. Bleeding gums was associated with higher SBP irrespective of periodontitis in the case control study and undetected hypertension was a common finding of this population of the UK. Lastly, periodontal treatment may assist in reducing the risk of hypertension, but based on the results herein observed, this effect remains inconclusive.

A previous systematic review assessed the epidemiological evidences on the link between periodontitis and hypertension (111). The authors concluded that periodontitis was associated to 50% increased odds for hypertension, which was greater (64%) for severe forms of the disease. The systematic review carried out for this PhD was designed to comprehensively investigate whether periodontitis as the independent variable would increase the odds of hypertension (outcome) and included not only epidemiological data but also assessment of prevalence, mean blood pressure, biomarkers, and treatment outcomes, including also interventional trials.

Other recent cross-sectional studies reported on the association between periodontitis and hypertension. Pietropaoli and co-workers carried out an analysis of the NHANES 2009-2014 waves and observed that periodontitis also impacted on successful hypertensive treatment (112). Those participants with periodontitis undergoing hypertensive therapy had a mean SBP of 2.3 to 3 mm Hg higher than those without periodontitis (P<0.0001). A more recent analysis of the NHANES III used a survey-based propensity score matching, accounting for common risk factors in the model. The authors observed that SBP increased gradually when comparing healthy individuals to those with

gingivitis (+2 mm Hg, $P<0.001$)," stable periodontitis" (+ 5.2 mmHg, $P<0.001$), or "unstable periodontitis" (+7.3mmHg, $P<0.001$) (113). A previous study by Tsakos and co-workers also looked at the same database (but without using propensity score matching analysis) and concluded that gingival bleeding was the only measure of periodontal inflammation consistently associated with elevated SBP and risk of hypertension. Each 10% increase in full mouth bleeding score was associated to a 0.5 mm Hg increase of SBP (99). Buhlin and co-workers found a significant association between self-reported gingival bleeding to known cardiovascular diseases (OR = 1.60, $P=0.0017$) (114). Additionally, in agreement with these findings, in a different study, Pietropaoli and co-workers reported that compared with no inflammation, the influence of severe PISA (periodontal inflamed surface area) and gingival bleeding were associated respectively with a 43% and 32% higher risk of high/uncontrolled blood pressure (≥130/80 mmHg) and with 4- and 5-mm Hg higher SBP (115). Concurrently, the SoPHiAS study evaluated the association between periodontitis and hypertension in a representative sample of the Portuguese population (116). Compared to participants without periodontitis, those with the disease had 2.31 times higher odds of hypertension (≥140/90 mm Hg) in the logistic model, adjusted for sociodemographic confounder such as gender, smoking habits, and BMI. Moreover, a 15.0% of undetected hypertension was observed, of which 62.5% had periodontitis and it was observed that age was a mediator of this association in the model.

Gingival bleeding, considered the key element for the detection of gingivitis, is directly correlated to bacterial plaque accumulation and dependent of plaque control (117). Thus, gingival inflammation control with oral hygiene measures is essential for the primary prevention of periodontitis. Various studies have investigated the effect of oral hygiene status on hypertension and cardiovascular risk (118-120). These studies reported that individuals who do not brush their teeth on a daily basis were at a high risk of hypertension, diabetes, hypertriglyceridemia and higher values of systemic inflammatory markers such as CRP and fibrinogen, hence increasing cardiovascular risk. Indeed, greatest periodontal bacterial burden (highest tertile of *Aggregatibacter. actinomycetemcomitans, Porphiromonas gingivalis, Treponema forsythia* and *Treponema denticola*) was associated with periodontitis status and 9-mm Hg and 5-mm Hg higher SBP/DBP respectively, and 3 times higher odds for hypertension in the

prospective INVEST study (121). This evidence would suggest an independent correlation between putative pathogens linked to periodontitis, oral hygiene measures, and hypertension.

Effects of periodontal health on hypertension

Several studies included in the systematic review showed a reduction in mean blood pressure levels upon provision of periodontal therapy, nevertheless this improvement in blood pressure control was inconsistently reported. The differences in study methodology, selection of the main primary outcome in the studies, inclusion criteria for the participants, treatment modality delivered, presence/absence of systemic conditions, timing of the follow-up appointment and sources of bias may have at least in part explained the discrepancy in results among the studies.

An interventional study by Seinost and co-workers including 30 systemically healthy patients with severe periodontitis reported improvement in endothelial function (P=0.003) and reduction in CRP (P=0.026) subsequent to periodontal treatment plus adjunctive systemic antibiotics, but no changes were observed in mean SBP/DBP at the 3-month follow-up (122). Similarly, Tonetti and co-workers carried out a RCT on 120 patients with generalised severe periodontitis and otherwise systemically healthy demonstrated that intensive periodontal treatment (IPT) involving supra- and subgingival debridement with locally delivered antibiotics improved endothelial function (P=0.001) and E-selectin (P=0.03) at the 6-month follow-up. The authors also observed an acute increase in systemic biomarkers as well as blood pressure levels 1-day after treatment with reconstitution at 2 and 6 months of the baseline blood pressure readings (89). Two different RCT's by Higashi and co-workers reported non-statistically significant decrease in blood pressure levels 24 weeks after periodontal therapy and systemic antibiotics (93, 123). The 2008 study included individuals with periodontitis and either systemically healthy or with hypertension while the 2009 study included participants with periodontitis and coronary artery disease, and they showed that periodontal therapy reduced the levels of CRP and IL-6 and augmented acetylcholine-induced vasodilation in these participants with periodontitis with and without hypertension as well as for those with coronary artery disease. Similar findings were observed in two other studies set out to assess the effects

of periodontal treatment on systemic markers of inflammation, whereby blood pressure levels were not altered (124, 125). Contrarily, the other 5 interventional studies included in the systematic review showed a decreased in mean blood pressure values (94, 95, 126-128). D'Aiuto and co-workers carried out a RCT on 40 patients with generalized severe periodontitis and otherwise healthy. IPT plus locally delivered antibiotics produced a 7 ±3 mm Hg (P=0.0211) reduction in SBP when compared to subgingival instrumentation alone in just two months after therapy (95). A greater difference (14±5 mm Hg; P=0.0124) was reported in current smokers. Vidal and colleagues observed a significant reduction of 12.5 mm Hg and 10 mm Hg in SBP and DBP respectively at 6 months reassessment following a course of non-surgical treatment of periodontitis in 26 patients with refractory hypertension and generalised severe chronic periodontitis (94). A recent 6-month follow up study for 40 systemically healthy participants with periodontitis confirmed these results (126). Delivery of non-surgical periodontal therapy and systemic antibiotics (amoxicillin and metronidazole) reduced SBP from 119.8±14.6 to 116.9±15.1 mm Hg (P=0.04) and DBP from 74.9±11.8 to 73.1±10.6 (P=0.05) (126). A sensitivity analysis was carried out to understand the influence of the antibiotics usage, confirming the consistency of the results even when antibiotics were not used. The most recent RCT carried out by D'Aiuto and co-workers on 256 participants with type II diabetes and generalised severe periodontitis showed a non-significant reduction of SBP = 2.3 mm Hg; 95%CI (–2.6, 7.2) and of DBP = 1.2 95% CI (–1.7, 4.1) at 1-year follow-up of combined non-surgical and surgical treatment of severe periodontitis (92). Lastly, Zhou and co-workers conducted the only RCT included in the systematic review with mean blood pressure levels as the primary outcome following treatment of periodontitis (involving supra and subgingival debridement with adjunctive local antimicrobials) on 107 pre-hypertensive patients with periodontitis (127). A 12.57 mm Hg reduction in SBP (P<0.05) and of 9.65 mm Hg in DBP (P<0.05) was observed in the test versus the control group. These authors also reported a decreased in endothelial microparticles.

Recently, Montero and co-workers also evaluated the effects of periodontal therapy on systemic markers of inflammation in 63 patients with severe periodontitis and metabolic syndrome (128). They carried out IPT (scaling and root planing with adjunctive systemic azithromycin) versus CPT (supragingival debridement with placebo) and

observed a of 7.3 mm Hg reduction in SBP (P = 0.008) and of 11.0 mm Hg in DBP (P=0.009) at the 6-month assessment.

A new systematic review and meta-analysis appraising the evidence on the effects of intensive periodontal treatment (subgingival instrumentation) compared to conventional treatment (supragingival scaling only/no treatment), with changes on blood pressure levels as the primary outcome (129). Eight RCTs met the inclusion criteria, comprising a variety of populations with different systemic conditions (from systemically healthy to refractory hypertension) and different lengths of follow-up (varying from 2 to 12 months). The main meta-analysis revealed a WMD of -4.3 mmHg (95%CI: -9.10–0.48, P=0.08) for SBP and of -3.16 mm Hg (95%CI: -6.51–0.19, P=0.06) for DBP, displaying high heterogeneity among the studies. Interestingly, the sensitivity analysis including only populations with either pre-hypertension or refractory hypertension (3 studies with various follow-up times) showed a statistically significant reduction in SBP of -11.41 mm Hg (P<0.00001) and of -8.43 mmHg (P<0.00001) in DBP, with low heterogeneity. Additionally, this review reported an improvement of endothelial function subsequently to periodontal treatment [FMD=1.49% (95% CI 0.9–2.08, p < 0.0001)], and significant reductions of CRP at all time points compared with control, but no changes were detected in other anti-inflammatory cytokines or lipids (129).

A number of limitations of the studies discussed above were identified, including the scarce number of studies carried out with a robust methodology reporting the effects of treatment of periodontitis on blood pressure levels and, particularly of those with reduction in blood pressure levels as the primary outcome. Nevertheless, the magnitude of the reduction in blood pressure levels observed following treatment of periodontitis is comparable to that observed when pharmacological and non-pharmacological interventions (such as weight loss, salt reduction, or regular aerobic exercise) have been assessed (30, 130-132). These intriguing findings could translate into important changes in clinical practice. Taken together, the studies reviewed in this PhD suggest that successful treatment of periodontitis could support the management of hypertension and ultimately result in cardiovascular risk reduction through significant blood pressure levels lowering, possibly via decreasing systemic inflammatory biomarkers such as CRP, and

improvement in endothelial function, particularly of those with levels of blood pressure within the currently accepted guidelines for hypertension definitions (i.e. SBP ≥ 130/140 and DBP ≥ 80/90 mm Hg) (129, 133).

The effects of a reduction of CRP and blood pressure levels on cardiovascular risks have been comprehensively assessed. The CANTOS study evaluated the benefits of canakinumab, a monoclonal antibody targeting up-stream inflammatory pathways to achieve a target of CRP levels <2mg/L in patients with excess cardiovascular risk. A 31% decrease in mortality (all causes) and cardiovascular mortality was observed in those participants who achieved a reduction of systemic inflammation based on the prespecified target (134). Other studies assessed the effects in cardiovascular risk reduction of anti-inflammatory drugs (colchicine in the COLCOT) and low dose immunosuppressants (methotrexate in the CIRT study) (135, 136). While the COLCOT study observed a significant reduction of ischemic cardiovascular event, the CIRT study failed to obtain a reduction in CRP or other inflammatory markers with no vascular benefit and greater side effects. This evidence would suggest that targeting inflammation could result in successful reduction of cardiovascular risk beyond optimal cardiovascular risk management (diet, cholesterol, blood pressure and so forth). A tight blood pressure control however involving both pharmacological and non-pharmacological strategies as well as precise assessment of the individual cardiovascular risk is essential to reduce cardiovascular illnesses and overall fatality in individuals with hypertension (137).

Confirmation that successful management of periodontitis could improve systemic health outcomes and decrease blood pressure levels needs further investigation in future larger multicentre studies assessing the true benefits of effective periodontal treatment and maintenance of periodontal health on hard cardiovascular outcomes including in patients with hypertension.

Causal link

The epidemiological and interventional evidence gathered and evaluated as part of this PhD supports a consistent association between periodontitis and hypertension.

Nevertheless, to ascertain whether this relationship is casual in nature, adopting recognized criteria and processes could prove useful (138). Ultimately, performing randomised controlled trials could produce unbiased evidence on the causal nature of the association between periodontitis and hypertension. Over the last 15 years a similar but more sophisticated approach named mendelian randomization (MR) surfaced and has further been validated as a tool to ascertain the causal nature of exposure-outcome associations (139).

In a recent investigation, data from these two experimental complementary approaches to test the causal association between periodontitis and hypertension was reported. A MR analysis and a 2-month follow-up RCT were performed. MR assessed single nucleotide polymorphisms (SNPs) linked to periodontitis in participants of the blood pressure Genome-Wide Association studies from the UK Biobank/International Consortium. A single-centre 2-month RCT included 101 hypertensive patients with periodontitis and evaluated the effects of intensive periodontal treatment on blood pressure levels as the primary outcome (140). Four SNPs in loci SIGLEC5, DEFA1A3, MTND1P5, and LOC107984137 (a proxy of periodontitis status) were linked to BP phenotypes. Additionally, at the 2-month follow-up the results from the interventional study revealed that intensive periodontal treatment resulted in a statistically significant reduction in SBP (-11.1 mm Hg; P<0.001)/DBP (8.3 mm Hg; P<0.001), endothelial function improvement, and reduction in inflammatory cytokines (IFN-c, IL-6, IL-17A, and TNF-a), and activated T-cell subsets when compared to participants in the control group. These results taken together would strongly suggest a causal link between periodontitis and hypertension. Nevertheless, this study is not exempt from limitations inherent to its design: on the one side, the specific genomic loci where heritability in periodontitis lies are still uncertain and hence it is unknown which SNPs are reliable proxy for periodontitis (141). On the other side, further studies are required to determine whether the BP reduction achieved at the 2-month evaluation is sustained beyond this short term follow-up.

Mechanisms supporting this association

Several animal models and clinical studies have shed light on the plausible biological mechanisms that identify periodontitis as a possible risk factor for hypertension. It is acknowledged that inflammation derived by specific infections pose an additional risk for vascular inflammation and atherosclerosis (142, 143). Periodontitis, originated by predominantly Gram-negative bacteria, has been recently recognised as one of the possible extravascular inflammatory conditions that influences vascular risk (143). Therefore, it has been hypothesised that bacteraemia from oral microbiota and the periodontitis-derived immune-inflammatory burden mediate vascular dysfunction and atherosclerotic inflammation increasing the risk of hypertension through structural and functional vasculature changes.

Aoyama and co-workers reported highly elevated periodontopathic bacteria (*A. Actinomycetemcomitans* and *Prevotella Intermedia*) from saliva and subgingival samples and their serum antibody titers in male patients with hypertension when compared to normotensive individuals or females (144).

Periodontitis-derived elevation of systemic inflammatory markers such as CRP, IL1, IL6, IL17, TNF- α, and immunological dysfunction of different cells involved in the adaptative, and immune systems have been linked to the pathogenies of hypertension. Evidence from an experimental (rat animal model) and a human study on periodontal inflammation confirmed that it would trigger an exacerbated production of PMN in the bone marrow, with primed innate immunity amplifying the inflammatory response in the event of a subsequent infection (145). The authors described it as one of the mechanisms linking periodontitis with other systemic diseases. Immune activation to Porphyromonas gingivalis was observed in another rat animal study, whereby an elicited Th1 immune response to this periodontal pathogen led to vascular inflammation, endothelial dysfunction and a subsequent rise in blood pressure (146). A cohort study using computer imaging [18 F-fluorodeoxyglucose positron emission tomography/computed tomography (18F-FDG-PET/CT)] presented another possible pathway linking periodontal and arterial vascular inflammation through boosting haematopoietic progenitor cells in the bone

marrow (147). These authors observed a strong link between periodontitis and arterial inflammation, which was mediated by increased hematopoietic activity.

Inflammatory processes can lead to the expression of adhesion molecules such as ICAM-1 and VCAM-1 by endothelial cells, which attract infiltration of leukocytes to the vasculature and are recognised markers for cardiovascular events involved in the pathology of hypertension (148). An animal model on ligature-induced periodontal inflammation revealed that periodontitis can also trigger activation and adhesion of monocytes/macrophages to endothelial cells, as observed through aortic activation of nuclear factor-κB, and VCAM-1 and TNF-α expression, which further amplify vascular inflammation (149).

Oxidative stress has also been associated to the pathogenesis of cardiovascular diseases and hypertension among other systemic conditions (150). Inflammation and oxidative stress act synergistically contributing to renal and vascular derangements in hypertension (151, 152). Oxidative stress, with high levels of ROS and activation of enzymatic processes by neutrophils has also a crucial role in the pathogenesis of periodontitis, favouring periodontal tissue break down (153, 154). Moreover, periodontal diseases can also be a source of systemic oxidative stress. An experimental rat animal study using the ligature-induced periodontal model showed increased serum levels of reactive oxygen species and reduced NOS-3 (antioxidant molecule) expression (155). D'Aiuto and co-workers in a case-control and interventional study observed that patients with generalised severe periodontitis displayed higher serum levels of Diacron-reactive oxygen metabolites (D-ROM) and lower total anti-oxidant capacity than controls with an elevation of ROS and inflammatory markers shortly after periodontal treatment (108). Additionally, a recent systematic review on the impact of periodontal therapy on oxidative stress markers including mostly non-RCT in predominantly systemically healthy populations and with a varying follow-up period (6 weeks-6 months) showed a reduction of oxidative stress levels (8-hydroxydeoxyguanosine) and total antioxidant status increase after the periodontal therapy (as assessed in GCF, saliva and serum), which was comparable to those without periodontitis (156).

In addition to the redox imbalance in ROS, disturbances in reactive nitrogen species (NOS) such as nitric oxide (NO) have also been involved in vascular pathology (157). NO, produced by endothelial nitric oxide synthase (eNOS), is a free radical potent vasodilator which has an effect in blood pressure control through regulation of vascular tone in the arterial walls. Homeostatic regulation of NO signalling is essential to avoid cardiovascular imbalance, whereby an excess or loss of NO bioavailability can derive in endothelial dysfunction (151, 158, 159). Endothelial dysfunction promotes reduction of NO availability, which has been strongly related to hypertension (160). It has been postulated that NO homeostasis can be affected by the entero-salivary nitrate-nitrite-nitric oxide pathway by nitrate-reducing bacteria, with an important role of the oral microbiota in the pathogenesis of hypertension through alterations in nitrite and NO formation (161, 162). A few studies have observed that using chlorhexidine mouthwashes that alter/eliminate nitrate-reducing bacteria in normotensive individuals with oral health led to SBP raise (163). Increased levels of NO have been reported in saliva of patients with periodontitis compared to periodontally healthy controls in some reports (164, 165), while others have described the opposite (166). Differences in the NO assessment method and factors influencing the methodology may be responsible for these differences. It seems that periodontal bacteria can affect NO availability and cause redox imbalances, which could represent another important mechanism justifying the increased BP levels observed in patients with periodontitis.

A series of experimental studies have argued another possible route whereby gut dysbiosis is influenced by periodontal bacteria (through translocation from the oral to the gut compartment), stimulating metabolic endotoxemia with an increased on systemic biomarkers and metabolic derangement leading to systemic conditions (167-170).

These experimental animal models and clinical studies above provided further insight to the epidemiological research findings and sum up current evidence supporting plausible mechanistic pathways linking periodontitis and hypertension. Additional studies are however warranted to further elucidate the exact mechanisms behind the association of periodontitis with blood pressure, with a focus on therapeutic approaches for the control of hypertension, periodontitis, and their complications.

Reverse causality

This PhD focused on elucidating the relationship between periodontitis and hypertension, with periodontitis as the independent variable and hypertension as the outcome variable. Nevertheless, it is acknowledged that the reverse might also possible, with hypertension conducive to periodontal inflammation.

A study in rats with renovascular induced hypertension observed vascular changes in gingival tissues and less often in the periodontal ligament with proliferation of the intima thickness, elastic layers and reduction of the lumen (171). Other two studies showed that compared with normotensive rats, increased bone loss and exacerbated periodontitis was observed in naturally hypertensive rats with experimentally induced periodontitis (172, 173). Another experimental study in spontaneously hypertensive rats compared to normotensive animals with induced periodontitis observed elevated serum CRP and gingival neutrophils, TNF- α and higher level of induced nitric oxide synthase in the gingival tissues, suggesting that hypertension favours the gingival inflammatory process (174). It has been also hypothesised that damage of small arterioles in hypertension could potentially increase gingival bleeding beyond periodontal diseases. Alterations of the renin-angiotensin aldosterone (RAS) system in the gingival tissues may have an impact on periodontitis and vice versa. A rat animal model study showed that Angiotensin II and other peptides were produced in the gingival vasculature (175). A subsequent experimental study in rats and humans suggested that adjustments in the RAS in the gingival tissues could prevent progression of bone loss (176). Based on this experimental evidence could be deduced therefore that the association between periodontitis and hypertension could be bi-directional in nature with each disease affecting the other and viceversa. Further research with pre-specified hypothesis should be performed to confirm or dispute these associations.

Strengths and Limitations

This doctoral thesis presented three comprehensive and complementary studies designed to elucidate the association between periodontitis and hypertension. A number

of strengths and limitations of each study design are stressed in all the three manuscripts herein presented.

Commonly, heterogeneity was observed in case definitions and study populations for both, periodontitis, and hypertension. Difficulties in establishing periodontitis and hypertension diagnosis were confronted, which made it difficult to combine the data. Another limitation in all three research methodologies was the inability to account for other hypertension risk factors such as diet rich in salt, abdominal obesity, use of anti-inflammatory medication, hormone treatments or immunosuppressants, and measurements of stress levels among others. Nevertheless, efforts were made to maintain consistency and robustness in each research methodology and to minimise bias. Several sensitivity analyses were performed in the three study designs to account for relevant factors and to minimise confounding.

The research question for this PhD was set out to elucidate the association between periodontitis and hypertension including different methodologies. The findings were grounded on the appraisal of observational/interventional evidence (the systematic review) and on the analysis of previously existing data retrieved from: i. two large databases of population-based surveys (the cross-sectional study) and ii. a newly created database containing baseline information obtained from previously conducted interventional trials (the case-control study). It is nonetheless acknowledged that study designs involving cross-sectional, case-control and sub-analysis of previously existing data are subject to bias and encompass limitations insofar ascertaining causality (177). Nonetheless, the findings of this doctoral thesis provided evidence that strengthened periodontitis could be an overlooked risk factor for hypertension, identifying the basis for further research on the topic.

Conclusions

5. CONCLUSIONS

Based on the findings from the studies conducted for this PhD, the following conclusions can be inferred:

1. Periodontitis is associated with elevated blood pressure levels and an increased risk of hypertension.
2. A linear association was observed: the more severe the periodontitis, the higher is the risk of hypertension.
3. The effects of periodontal therapy upon reduction of SBP/DBP remains inconclusive.
4. Periodontitis is linked to low-grade systemic inflammation (measured as CRP levels and WBC counts).
5. Low-grade systemic inflammation could be a key mediator of the association.
6. A greater proportion of undetected hypertension was observed in patients with periodontitis compared to those without periodontitis.
7. Gingival inflammation per se is associated to elevated SBP.
8. Longer and larger studies are needed however to determine whether periodontal treatment leading to a decrease in systemic inflammation, may represent a novel nonpharmacologic intervention in hypertension management that benefit patients in terms of CV health, ultimately resulting in reduced morbidity and mortality. Oral health professionals could play a pivotal role in helping the medical community detecting and tackling the burden and consequences of hypertension.

6. CONCLUSIONS IN SPANISH

Las siguientes conclusiones se derivan de los estudios llevados a cabo en esta tesis doctoral:

1. La periodontitis está asociada con niveles elevados de presión arterial y con un mayor riesgo de hipertensión.
2. Se ha observado una relación lineal: cuanto más severa es la periodontitis, mayor es el riesgo de hipertensión.
3. El tratamiento periodontal no ha conseguido demostrar una reducción de la presión arterial de manera consistente.
4. La periodontitis está asociada a una elevación de los marcadores de inflamación sistémica (proteína C-reactiva y recuento de glóbulos blancos).
5. La inflamación sistémica de bajo grado podría actuar como un mediador clave de la asociación.
6. Se observó una mayor proporción de casos de hipertensión arterial no diagnosticada en pacientes con enfermedad periodontal en comparación con aquellos sin periodontitis.
7. La inflamación gingival por si misma está asociada a la PAS.
8. Se necesitan estudios con una mayor muestra y tiempo de seguimiento para determinar si el tratamiento periodontal, a través de una reducción de los marcadores de inflamación sistémica, podría representar una intervención no farmacológica en el control de la hipertensión, beneficiando la salud cardiovascular y con el fin último de reducir morbilidad y mortalidad. Los profesionales de la salud oral podrían jugar un papel crucial ayudando a la comunidad médica a detectar y controlar la hipertensión y sus consecuencias.

Clinical implications and further research

7. CLINICAL IMPLICATIONS AND FURTHER RESEARCH

Successful clinical approaches to tackle periodontitis and hypertension would depend largely on raising awareness among physicians, oral health professionals, policy makers and the population as a whole in order to inform about this association and its common risk factors. These highly widespread conditions are generally preventable and prompt recognition is essential for effective therapies.

Periodontitis remains poorly diagnosed among dental professionals, which has been one of the most common causes of professional litigation (178). Training the dental community in the screening and management of periodontal diseases has been the focus of the periodontal societies worldwide for several years. Similarly, several studies have shown that the medical community has varying levels of understanding of periodontal diseases and their effects beyond the oral cavity, pointing towards an overall need for further continuing medical education courses and involvement in multidisciplinary teams (179-181). A very recent study evaluated the validity of a predictive model for screening of periodontitis, based on risk factors such as age, gender, ethnicity, HbA1c and smoking habit, from the NHANES 2010-2012 database and suggested this could be used as a reliable screening tool for periodontitis in primary medical care settings with the purpose to facilitate referral to the dental setting of patients with a suspected periodontal diagnosis (182). Early diagnosis and management of gingivitis and periodontitis is essential to achieve oral health as this is amenable to prevention (183). A recent analysis of the economic burden of periodontitis in 2018 confirmed that it produced a loss of €149.52B and €122.65B in Europe and the USA, respectively (184). Data from a medical insurance database of the Dutch population showed that effective prevention and treatment of periodontitis was a very cost-effective treatment in managing medical related costs such as those from diagnosis, treatment and hospitalization in patients with type II diabetes (185). A further study completed using the NHANES 2009-2014 cross-sectional waves suggested that the treatment of periodontitis could be a cost-effective strategy in patients with type-II diabetes as to reduce oral health diseases burden with a reduction of tooth loss but also of microvascular complications and CVD in the US population (186).

Early detection of high blood pressure through screening programs has been suggested as a valuable tool in identifying those at risk of hypertension, leading to both preventive measures and early management to reduce morbidity and mortality and ultimately assisting to curve the social and economic burden of elevated blood pressure in any health system worldwide (2). Due to the high levels of undiagnosed hypertension observed in this PhD study, we suggest that development and implementation of hypertension screening systems in dental settings could be a valuable asset in assisting the management of hypertension. Hypertension screening should be performed with trained professionals, appropriate equipment, patient education and informed consent, along with good relationships between health professions (2). It is acknowledged that a white coat effect in dental settings could represent a major drawback for obtaining accurate blood pressure measurements (187). In order to overcome this limitation and to improve the validity of the screening methods by oral health professionals, it is important to develop hypertension screening protocols that include recognition of common risk factors, use of simple validated questionnaires and optimization of the blood pressure measurements (188). The following proposals could be considered: a) adopting current clinical guidelines for hypertension management (189), b) using standardized oscillometer devices for unsupervised blood pressure measurement (190) and c) set an appropriate timing for the recording during the dental visit where the patient is not anxious and local anaesthesia has not been administered (for instance at the end of a baseline clinical consultation).

Oral health professionals could play a pivotal role in providing guidance not only oral health advice but also information for common risk factors such as to maintain a healthy and balanced diet with sugar, salt and fat restriction, smoking cessation, promoting regular physical activity, alcohol consumption reduction, and diabetes control (191).

Further research should focus on a) the pathogenetic biological mechanisms that shape the relationship between hypertension and periodontitis, b) investigating the appropriateness and validity of hypertension screening in the dental setting and c). evaluating the impact and cost-effectiveness of the treatment of periodontitis as a novel

non-pharmacological intervention to reduce the risk of hypertension and related complications through well-powered randomised clinical trials.

From a public health perspective, treatment of periodontitis and its possible effects on hypertension prevention and management can have profound implications for public health policies, including attention to the inclusion of oral health strategies in current systemic health schemes.

Bibliography

8. BIBLIOGRAPHY

1. Walker HK, Hall WD, Hurst JW. Clinical methods: the history, physical, and laboratory examinations: Butterworth-heinemann; 1990.
2. Organization WH. A global brief on hypertension: silent killer, global public health crisis: World Health Day 2013. World Health Organization; 2013.
3. Chobanian AV, Bakris GL, Black HR, Cushman WC, Green LA, Izzo Jr JL, et al. The seventh report of the joint national committee on prevention, detection, evaluation, and treatment of high blood pressure: the JNC 7 report. Jama. 2003;289:2560-71.
4. Mancia G, Fagard R, Narkiewicz K, Redon J, Zanchetti A, Böhm M, et al. 2013 ESH/ESC guidelines for the management of arterial hypertension: the Task Force for the Management of Arterial Hypertension of the European Society of Hypertension (ESH) and of the European Society of Cardiology (ESC). Blood pressure. 2013;22:193-278.
5. Hodgkinson J, Mant J, Martin U, Guo B, Hobbs F, Deeks J, et al. Relative effectiveness of clinic and home blood pressure monitoring compared with ambulatory blood pressure monitoring in diagnosis of hypertension: systematic review. Bmj. 2011;342.
6. Muntner P, Shimbo D, Carey RM, Charleston JB, Gaillard T, Misra S, et al. Measurement of blood pressure in humans: a scientific statement from the American Heart Association. Hypertension. 2019;73(5):e35-e66.
7. Williams B, Mancia G, Spiering W, Agabiti Rosei E, Azizi M, Burnier M, et al. 2018 ESC/ESH Guidelines for the management of arterial hypertension: The Task Force for the management of arterial hypertension of the European Society of Cardiology (ESC) and the European Society of Hypertension (ESH). European heart journal. 2018;39(33):3021-104.
8. Whelton PK, Carey RM, Aronow WS, Casey DE, Collins KJ, Himmelfarb CD, et al. 2017 ACC/AHA/AAPA/ABC/ACPM/AGS/APhA/ASH/ASPC/NMA/PCNA guideline for the prevention, detection, evaluation, and management of high blood pressure in adults: a report of the American College of Cardiology/American Heart Association Task Force on Clinical Practice Guidelines. Journal of the American College of Cardiology. 2018;71(19):e127-e248.
9. Bakris G, Ali W, Parati G. ACC/AHA versus ESC/ESH on hypertension guidelines: JACC guideline comparison. Journal of the American College of Cardiology. 2019;73(23):3018-26.
10. Zhou B, Bentham J, Di Cesare M, Bixby H, Danaei G, Cowan MJ, et al. Worldwide trends in blood pressure from 1975 to 2015: a pooled analysis of 1479 population-based measurement studies with 19· 1 million participants. The Lancet. 2017;389(10064):37-55.
11. Chow CK, Teo KK, Rangarajan S, Islam S, Gupta R, Avezum A, et al. Prevalence, awareness, treatment, and control of hypertension in rural and urban communities in high-, middle-, and low-income countries. Jama. 2013;310(9):959-68.
12. Muntner P, Abdalla M, Correa A, Griswold M, Hall JE, Jones DW, et al. Hypertension in blacks: unanswered questions and future directions for the JHS (Jackson Heart Study). Hypertension. 2017;69(5):761-9.

13. Carretero OA, Oparil S. Essential hypertension part I: definition and etiology. Circulation. 2000;101:329-35.
14. Beevers G, Lip GYH, O'Brien E. ABC of hypertension: the pathophysiology of hypertension. Br Med J. 2001;322:912.
15. Luft FC. Twins in cardiovascular genetic research. Hypertension. 2001;37:350-6.
16. Evangelou E, Warren HR, Mosen-Ansorena D, Mifsud B, Pazoki R, Gao H, et al. Genetic analysis of over 1 million people identifies 535 new loci associated with blood pressure traits. Nature genetics. 2018;50(10):1412-25.
17. Mills KT, Stefanescu A, He J. The global epidemiology of hypertension. Nature Reviews Nephrology. 2020:1-15.
18. Joffres M, Falaschetti E, Gillespie C, Robitaille C, Loustalot F, Poulter N, et al. Hypertension prevalence, awareness, treatment and control in national surveys from England, the USA and Canada, and correlation with stroke and ischaemic heart disease mortality: a cross-sectional study. BMJ open. 2013;3(8):e003423.
19. Dinh QN, Drummond GR, Sobey CG, Chrissobolis S. Roles of inflammation, oxidative stress, and vascular dysfunction in hypertension. BioMed research international. 2014;2014.
20. Dzielak DJ. The immune system and hypertension. Hypertension. 1992;19(1_supplement):I36.
21. Wenzel UO, Ehmke H, Bode M. Immune mechanisms in arterial hypertension. Recent advances. Cell and Tissue Research.1-12.
22. Rodríguez-Iturbe B, Pons H, Quiroz Y, Gordon K, Rincón J, Chávez M, et al. Mycophenolate mofetil prevents salt-sensitive hypertension resulting from angiotensin II exposure. Kidney international. 2001;59:2222-32.
23. Madhur MS, Lob HE, McCann LA, Iwakura Y, Blinder Y, Guzik TJ, et al. Interleukin 17 promotes angiotensin II–induced hypertension and vascular dysfunction. Hypertension. 2010;55:500-7.
24. Kampus P, Muda P, Kals J, Ristimäe T, Fischer K, Teesalu R, et al. The relationship between inflammation and arterial stiffness in patients with essential hypertension. International journal of cardiology. 2006;112(1):46-51.
25. Kim YM, Kim MY, Kim HJ, Roh GS, Ko GH, Seo HG, et al. Compound C independent of AMPK inhibits ICAM-1 and VCAM-1 expression in inflammatory stimulants-activated endothelial cells in vitro and in vivo. Atherosclerosis. 2011;219:57-64.
26. Drummond GR, Vinh A, Guzik TJ, Sobey CG. Immune mechanisms of hypertension. Nature Reviews Immunology. 2019;19(8):517-32.
27. Lim SS, Vos T, Flaxman AD, Danaei G, Shibuya K, Adair-Rohani H, et al. A comparative risk assessment of burden of disease and injury attributable to 67 risk factors and risk factor clusters in 21 regions, 1990–2010: a systematic analysis for the Global Burden of Disease Study 2010. The lancet. 2012;380(9859):2224-60.
28. Lewington S. Prospective studies collaboration. Age-specific relevance of usual blood pressure to vascular mortality: a meta-analysis of individual data for one million adults in 61 prospective studies. Lancet. 2002;360:1903-13.

29. Forouzanfar MH, Liu P, Roth GA, Ng M, Biryukov S, Marczak L, et al. Global burden of hypertension and systolic blood pressure of at least 110 to 115 mm Hg, 1990-2015. Jama. 2017;317(2):165-82.
30. Ettehad D, Emdin CA, Kiran A, Anderson SG, Callender T, Emberson J, et al. Blood pressure lowering for prevention of cardiovascular disease and death: a systematic review and meta-analysis. The Lancet. 2016;387(10022):957-67.
31. Group SR. A randomized trial of intensive versus standard blood-pressure control. New England Journal of Medicine. 2015;373(22):2103-16.
32. Bartold PM, Van Dyke TE. Periodontitis: a host-mediated disruption of microbial homeostasis. Unlearning learned concepts. Periodontol 2000. 2013;62:203-17.
33. Hajishengallis G. Immunomicrobial pathogenesis of periodontitis: keystones, pathobionts, and host response. Trends Immunol. 2014;35(1):3-11.
34. Tonetti MS, Greenwell H, Kornman KS. Staging and grading of periodontitis: Framework and proposal of a new classification and case definition. Journal of periodontology. 2018;89:S159-S72.
35. Kassebaum NJ, Bernabé E, Dahiya M, Bhandari B, Murray CJL, Marcenes W. Global Burden of Severe Periodontitis in 1990-2010 A Systematic Review and Meta-regression. J Dent Res. 2014:0022034514552491.
36. Frencken JE, Sharma P, Stenhouse L, Green D, Laverty D, Dietrich T. Global epidemiology of dental caries and severe periodontitis–a comprehensive review. J Clin Periodontol. 2017;44(S18).
37. Kassebaum N, Kyu HH, Zoeckler L, Olsen HE, Thomas K, Pinho C, et al. Child and adolescent health from 1990 to 2015: findings from the global burden of diseases, injuries, and risk factors 2015 study. JAMA pediatrics. 2017;171(6):573-92.
38. Genco RJ, Borgnakke WS. Risk factors for periodontal disease. Periodontol 2000. 2013;62(1):59-94.
39. Eke PI, Dye BA, Wei L, Slade GD, Thornton-Evans GO, Borgnakke WS, et al. Update on prevalence of periodontitis in adults in the United States: NHANES 2009 to 2012. J Periodontol. 2015;86:611-22.
40. Sabbah W, Tsakos G, Sheiham A, Watt RG. The effects of income and education on ethnic differences in oral health: a study in US adults. J Epidemiol Community Health. 2009;63(7):516-20.
41. Borges TdF, Regalo SC, Taba Jr M, Siéssere S, Mestriner Jr W, Semprini M. Changes in masticatory performance and quality of life in individuals with chronic periodontitis. Journal of periodontology. 2013;84(3):325-31.
42. Buset S, Walter C, Friedmann A, Weiger R, Borgnakke WS, Zitzmann NU. Are periodontal diseases really silent? A systematic review of their effect on quality of life. J Clin Periodontol. 2016.
43. Ferreira M, Dias-Pereira A, Branco-de-Almeida L, Martins C, Paiva S. Impact of periodontal disease on quality of life: a systematic review. Journal of periodontal research. 2017;52(4):651-65.
44. Baehni P, Tonetti MS. Conclusions and consensus statements on periodontal health, policy and education in Europe: a call for action–consensus view 1. Eur J Dent Educ. 2010;14:2-3.

45. Tonetti MS, Jepsen S, Jin L, Otomo-Corgel J. Impact of the global burden of periodontal diseases on health, nutrition and wellbeing of mankind: A call for global action. J Clin Periodontol. 2017;44(5):456-62.
46. Löe H, Theilade E, Jensen SB. Experimental gingivitis in man. J Periodontol. 1965;36:177-87.
47. Flemmig TF. Periodontitis. Annals of Periodontology. 1999;4(1):32-7.
48. Socransky SS, Haffajee AD, Cugini MA, Smith C, Kent RL. Microbial complexes in subgingival plaque. J Clin Periodontol. 1998;25:134-44.
49. Al-Yahfoufi Z, Mombelli A, Wicki A, Lang NP. The effect of plaque control in subjects with shallow pockets and high prevalence of periodontal pathogens. J Clin Periodontol. 1995;22(1):78-84.
50. Bartold PM, Van Dyke TE. An appraisal of the role of specific bacteria in the initial pathogenesis of periodontitis. Wiley Online Library; 2019.
51. Hajishengallis G, Lamont RJ. Beyond the red complex and into more complexity: the polymicrobial synergy and dysbiosis (PSD) model of periodontal disease etiology. Molecular oral microbiology. 2012;27:409-19.
52. Lamont RJ, Koo H, Hajishengallis G. The oral microbiota: dynamic communities and host interactions. Nature Reviews Microbiology. 2018;16(12):745-59.
53. Hajishengallis G. Periodontitis: from microbial immune subversion to systemic inflammation. Nature Reviews Immunology. 2015;15:30-44.
54. Kornman KS. Mapping the pathogenesis of periodontitis: a new look. J Periodontol. 2008;79:1560-8.
55. Ebersole JL, Taubman MA. The protective nature of host responses in periodontal diseases. Periodontol 2000. 1994;5:112-41.
56. Ebersole JL, Dawson DR, Morford LA, Peyyala R, Miller CS, Gonzaléz OA. Periodontal disease immunology:‘double indemnity’in protecting the host. Periodontol 2000. 2013;62:163-202.
57. Lang NP, Lindhe J. Clinical periodontology and implant dentistry. 2015.
58. Serhan CN, Chiang N, Van Dyke TE. Resolving inflammation: dual anti-inflammatory and pro-resolution lipid mediators. Nature Reviews Immunology. 2008;8(5):349.
59. Freire MO, Van Dyke TE. Natural resolution of inflammation. Periodontol 2000. 2013;63(1):149-64.
60. Page RC, Schroeder HE. Pathogenesis of inflammatory periodontal disease. A summary of current work. Laboratory investigation; a journal of technical methods and pathology. 1976;34:235-49.
61. Offenbacher S. Periodontal diseases: pathogenesis. Ann Periodontol. 1996;1(1):821-78.
62. Reynolds JJ, Meikle MC. Mechanisms of connective tissue matrix destruction in periodontitis. Periodontology 2000. 1997;14(1):144-57.
63. Mogi M, Otogoto J, Ota N, Togari A. Differential expression of RANKL and osteoprotegerin in gingival crevicular fluid of patients with periodontitis. Journal of dental research. 2004;83(2):166-9.
64. Kadowaki N, Ho S, Antonenko S, de Waal Malefyt R, Kastelein RA, Bazan F, et al. Subsets of human dendritic cell precursors express different toll-like receptors and

respond to different microbial antigens. The Journal of experimental medicine. 2001;194(6):863-70.
65. Seymour G, Gemmell E, Reinhardt RA, Eastcott J, Taubman M. Immunopathogenesis of chronic inflammatory periodontal disease: cellular and molecular mechanisms. Journal of periodontal research. 1993;28(7):478-86.
66. Tsukasaki M, Komatsu N, Nagashima K, Nitta T, Pluemsakunthai W, Shukunami C, et al. Host defense against oral microbiota by bone-damaging T cells. Nature communications. 2018;9(1):1-11.
67. Dutzan N, Kajikawa T, Abusleme L, Greenwell-Wild T, Zuazo CE, Ikeuchi T, et al. A dysbiotic microbiome triggers TH17 cells to mediate oral mucosal immunopathology in mice and humans. Science translational medicine. 2018;10(463).
68. Hunter W. THE COMING OF AGE OF ORAL SEPSIS. British Medical Journal. 1921;1(3154):859-.
69. Monsarrat P, Blaizot A, Kémoun P, Ravaud P, Nabet C, Sixou M, et al. Clinical research activity in periodontal medicine: a systematic mapping of trial registers. Journal of Clinical Periodontology. 2016;43(5):390-400.
70. Sanz M, Marco del Castillo A, Jepsen S, Gonzalez-Juanatey JR, D'Aiuto F, Bouchard P, et al. Periodontitis and cardiovascular diseases: Consensus report. J Clin Periodontol. 2020;47(3):268-88.
71. Sanz M, Ceriello A, Buysschaert M, Chapple I, Demmer RT, Graziani F, et al. Scientific evidence on the links between periodontal diseases and diabetes: Consensus report and guidelines of the joint workshop on periodontal diseases and diabetes by the International Diabetes Federation and the European Federation of Periodontology. Diabetes Research and Clinical Practice. 2018;137:231-41.
72. Sanz M, Kornman K, Working Group 3 of the Joint EFPAAPW. Periodontitis and adverse pregnancy outcomes: consensus report of the Joint EFP/AAP Workshop on Periodontitis and Systemic Diseases. Journal of Periodontology. 2013;84:S164-S9.
73. Genco RJ, Sanz M. Clinical and public health implications of periodontal and systemic diseases: An overview. Periodontology 2000. 2020;83(1):7-13.
74. Hajishengallis G, Chavakis T. Local and systemic mechanisms linking periodontal disease and inflammatory comorbidities. Nature Reviews Immunology. 2021:1-15.
75. Kozarov EV, Dorn BR, Shelburne CE, Dunn Jr WA, Progulske-Fox A. Human atherosclerotic plaque contains viable invasive Actinobacillus actinomycetemcomitans and Porphyromonas gingivalis. Arteriosclerosis, thrombosis, and vascular biology. 2005;25(3):e17-e8.
76. Schenkein HA, Papapanou PN, Genco R, Sanz M. Mechanisms underlying the association between periodontitis and atherosclerotic disease. Periodontology 2000. 2020;83(1):90-106.
77. Paraskevas S, Huizinga JD, Loos BG. A systematic review and meta-analyses on C-reactive protein in relation to periodontitis. Journal of clinical periodontology. 2008;35:277-90.
78. Loos BG, Craandijk J, Hoek FJ, Dillen PMWv, Van Der Velden U. Elevation of systemic markers related to cardiovascular diseases in the peripheral blood of periodontitis patients. Journal of periodontology. 2000;71(10):1528-34.

79. Arimatsu K, Yamada H, Miyazawa H, Minagawa T, Nakajima M, Ryder MI, et al. Oral pathobiont induces systemic inflammation and metabolic changes associated with alteration of gut microbiota. Scientific reports. 2014;4(1):1-9.
80. Jepsen S, Deschner J, Braun A, Schwarz F, Eberhard J. Calculus removal and the prevention of its formation. Periodontology 2000. 2011;55(1):167-88.
81. Heitz-Mayfield LJ, Lang NP. Surgical and nonsurgical periodontal therapy. Learned and unlearned concepts. Periodontology 2000. 2013;62(1):218-31.
82. Tomasi C, Leyland AH, Wennström JL. Factors influencing the outcome of non-surgical periodontal treatment: a multilevel approach. Journal of clinical periodontology. 2007;34:682-90.
83. Lang NP, Tonetti MS. Periodontal risk assessment (PRA) for patients in supportive periodontal therapy (SPT). Oral Health Prev Dent. 2003;1:7-16.
84. Magnusson I, Lindhe J, Yoneyama T, Liljenberg B. Recolonization of a subgingival microbiota following scaling in deep pockets. Journal of clinical periodontology. 1984;11(3):193-207.
85. Ramfjord SP, Caffesse RG, Morrison EC, Hill RW, Kerry GJ, Appleberry EA, et al. 4 modalities of periodontal treatment compared over 5 years. Journal of clinical periodontology. 1987;14:445-52.
86. Simpson TC, Weldon JC, Worthington HV, Needleman I, Wild SH, Moles DR, et al. Treatment of periodontal disease for glycaemic control in people with diabetes mellitus. The Cochrane Library. 2015.
87. Bi WG, Emami E, Luo Z-C, Santamaria C, Wei SQ. Effect of periodontal treatment in pregnancy on perinatal outcomes: a systematic review and meta-analysis. The Journal of Maternal-Fetal & Neonatal Medicine. 2019:1-10.
88. D'Aiuto F, Orlandi M, Gunsolley JC. Evidence that periodontal treatment improves biomarkers and CVD outcomes. Journal of periodontology. 2013;84:S85-S105.
89. Tonetti MS, D'aiuto F, Nibali L, Donald A, Storry C, Parkar M, et al. Treatment of periodontitis and endothelial function. New England Journal of Medicine. 2007;356(9):911-20.
90. Caúla AL, Lira-Junior R, Tinoco EMB, Fischer RG. The effect of periodontal therapy on cardiovascular risk markers: a 6-month randomized clinical trial. Journal of clinical periodontology. 2014;41(9):875-82.
91. Freitas COTd, Gomes-Filho IS, Naves RC, Nogueira Filho GdR, Cruz SSd, Santos CAdST, et al. Influence of periodontal therapy on C-reactive protein level: a systematic review and meta-analysis. J. 2012;20(1):1-8.
92. D'Aiuto F, Gkranias N, Bhowruth D, Khan T, Orlandi M, Suvan J, et al. Systemic effects of periodontitis treatment in patients with type 2 diabetes: a 12 month, single-centre, investigator-masked, randomised trial. The Lancet Diabetes & Endocrinology. 2018;6(12):954-65.
93. Higashi Y, Goto C, Jitsuiki D, Umemura T, Nishioka K, Hidaka T, et al. Periodontal infection is associated with endothelial dysfunction in healthy subjects and hypertensive patients. Hypertension. 2008;51(2):446-53.

94. Vidal F, Cordovil I, Figueredo CMS, Fischer RG. Non-surgical periodontal treatment reduces cardiovascular risk in refractory hypertensive patients: a pilot study. Journal of clinical periodontology. 2013;40:681-7.
95. D'Aiuto F, Parkar M, Nibali L, Suvan J, Lessem J, Tonetti MS. Periodontal infections cause changes in traditional and novel cardiovascular risk factors: results from a randomized controlled clinical trial. American heart journal. 2006;151:977-84.
96. Jeffcoat MK, Jeffcoat RL, Gladowski PA, Bramson JB, Blum JJ. Impact of periodontal therapy on general health: evidence from insurance data for five systemic conditions. American journal of preventive medicine. 2014;47(2):166-74.
97. Shanbhag S, Dahiya M, Croucher R. The impact of periodontal therapy on oral health-related quality of life in adults: a systematic review. Journal of clinical periodontology. 2012;39:725-35.
98. Botelho J, Machado V, Proença L, Bellini DH, Chambrone L, Alcoforado G, et al. The impact of nonsurgical periodontal treatment on oral health-related quality of life: a systematic review and meta-analysis. Clinical Oral Investigations. 2020;24(2):585-96.
99. Tsakos G, Sabbah W, Hingorani AD, Netuveli G, Donos N, Watt RG, et al. Is periodontal inflammation associated with raised blood pressure? Evidence from a National US survey. Journal of hypertension. 2010;28:2386-93.
100. Tsioufis C, Kasiakogias A, Thomopoulos C, Stefanadis C. Periodontitis and blood pressure: the concept of dental hypertension. Atherosclerosis. 2011;219(1):1-9.
101. Del Pinto R, Pietropaoli D, Munoz-Aguilera E, D'Aiuto F, Czesnikiewicz-Guzik M, Monaco A, et al. Periodontitis and hypertension: is the association causal? High Blood Pressure & Cardiovascular Prevention. 2020;27:281-9.
102. Lockhart PB, Bolger AF, Papapanou PN, Osinbowale O, Trevisan M, Levison ME, et al. Periodontal disease and atherosclerotic vascular disease: does the evidence support an independent association? A scientific statement from the American Heart Association. Circulation. 2012;125:2520-44.
103. Türkoğlu O, Barış N, Kütükçüler N, Şenarslan Ö, Güneri S, Atilla G. Evaluation of serum anti-cardiolipin and oxidized low-density lipoprotein levels in chronic periodontitis patients with essential hypertension. Journal of periodontology. 2008;79(2):332-40.
104. Leong X-F, Ng C-Y, Badiah B, Das S. Association between hypertension and periodontitis: possible mechanisms. The Scientific World Journal. 2014;2014.
105. Androulakis ES, Tousoulis D, Papageorgiou N, Tsioufis C, Kallikazaros I, Stefanadis C. Essential hypertension: is there a role for inflammatory mechanisms? Cardiology in review. 2009;17(5):216-21.
106. Sesso HD, Buring JE, Rifai N, Blake GJ, Gaziano JM, Ridker PM. C-reactive protein and the risk of developing hypertension. Jama. 2003;290(22):2945-51.
107. Amar S, Gokce N, Morgan S, Loukideli M, Van Dyke TE, Vita JA. Periodontal disease is associated with brachial artery endothelial dysfunction and systemic inflammation. Arteriosclerosis, thrombosis, and vascular biology. 2003;23(7):1245-9.
108. D'aiuto F, Nibali L, Parkar M, Patel K, Suvan J, Donos N. Oxidative stress, systemic inflammation, and severe periodontitis. Journal of dental research. 2010;89(11):1241-6.

109. Lucia Macedo Paizan M, Fernando Vilela-Martin J. Is there an association between periodontitis and hypertension? Current cardiology reviews. 2014;10:355-61.
110. Tonetti MS, Dyke TE. Periodontitis and atherosclerotic cardiovascular disease: consensus report of the Joint EFP/AAP Workshop on Periodontitis and Systemic Diseases. Journal of clinical periodontology. 2013;40.
111. Martin-Cabezas R, Seelam N, Petit C, Agossa K, Gaertner S, Tenenbaum H, et al. Association between periodontitis and arterial hypertension: A systematic review and meta-analysis. American heart journal. 2016;180:98-112.
112. Pietropaoli D, Del Pinto R, Ferri C, Wright Jr JT, Giannoni M, Ortu E, et al. Poor oral health and blood pressure control among US hypertensive adults: results from the National Health and Nutrition Examination Survey 2009 to 2014. Hypertension. 2018;72(6):1365-73.
113. Pietropaoli D, Monaco A, D'Aiuto F, Aguilera EM, Ortu E, Giannoni M, et al. Active gingival inflammation is linked to hypertension. Journal of hypertension. 2020;38(10):2018-27.
114. Buhlin K, Gustafsson A, Håkansson J, Klinge B. Oral health and cardiovascular disease in Sweden. Journal of clinical periodontology. 2002;29(3):254-9.
115. Pietropaoli D, Del Pinto R, Ferri C, Marzo G, Giannoni M, Ortu E, et al. Association between periodontal inflammation and hypertension using periodontal inflamed surface area and bleeding on probing. Journal of clinical periodontology. 2020;47(2):160-72.
116. Machado V, Aguilera EM, Botelho J, Hussain SB, Leira Y, Proença L, et al. Association between periodontitis and high blood pressure: results from the study of periodontal health in Almada-Seixal (SoPHiAS). Journal of clinical medicine. 2020;9(5):1585.
117. Murakami S, Mealey BL, Mariotti A, Chapple IL. Dental plaque–induced gingival conditions. Journal of clinical periodontology. 2018;45:S17-S27.
118. Choi HM, Han K, Park YG, Park JB. Associations among oral hygiene behavior and hypertension prevalence and control: the 2008 to 2010 Korea National Health and Nutrition Examination Survey. Journal of periodontology. 2015;86(7):866-73.
119. de Oliveira C, Watt R, Hamer M. Toothbrushing, inflammation, and risk of cardiovascular disease: results from Scottish Health Survey. Bmj. 2010;340.
120. Fujita M, Ueno K, Hata A. Lower frequency of daily teeth brushing is related to high prevalence of cardiovascular risk factors. Experimental biology and medicine. 2009;234(4):387-94.
121. Desvarieux M, Demmer RT, Jacobs Jr DR, Rundek T, Boden-Albala B, Sacco RL, et al. Periodontal bacteria and hypertension: the oral infections and vascular disease epidemiology study (INVEST). Journal of hypertension. 2010;28(7):1413.
122. Seinost G, Wimmer G, Skerget M, Thaller E, Brodmann M, Gasser R, et al. Periodontal treatment improves endothelial dysfunction in patients with severe periodontitis. American heart journal. 2005;149(6):1050-4.
123. Higashi Y, Goto C, Hidaka T, Soga J, Nakamura S, Fujii Y, et al. Oral infection-inflammatory pathway, periodontitis, is a risk factor for endothelial dysfunction in patients with coronary artery disease. Atherosclerosis. 2009;206:604-10.

124. Taylor B, Tofler G, Morel-Kopp MC, Carey H, Carter T, Elliott M, et al. The effect of initial treatment of periodontitis on systemic markers of inflammation and cardiovascular risk: a randomized controlled trial. European journal of oral sciences. 2010;118(4):350-6.
125. Graziani F, Cei S, Tonetti M, Paolantonio M, Serio R, Sammartino G, et al. Systemic inflammation following non-surgical and surgical periodontal therapy. Journal of clinical periodontology. 2010;37(9):848-54.
126. Houcken W, Teeuw WJ, Bizzarro S, Rodriguez EA, Mulders TA, van den Born BJH, et al. Arterial stiffness in periodontitis patients and controls A case-control and pilot intervention study. Journal of Human Hypertension. 2016;30(1):24-9.
127. Zhou QB, Xia WH, Ren J, Yu BB, Tong XZ, Chen YB, et al. Effect of intensive periodontal therapy on blood pressure and endothelial microparticles in patients with prehypertension and periodontitis: a randomized controlled trial. Journal of periodontology. 2017;88(8):711-22.
128. Yang YL. Clinical efficacy and safety of urapidil injection on acute heart failure in elderly hypertensive patients complicated with diabetes mellitus. [Chinese]. Chinese Journal of New Drugs. 2017;26(15):1814-7.
129. Sharma S, Sridhar S, McIntosh A, Messow C-M, Aguilera EM, Del Pinto R, et al. Periodontal therapy and treatment of hypertension-alternative to the pharmacological approach. A systematic review and meta-analysis. Pharmacological Research. 2021;166:105511.
130. Neter JE, Stam BE, Kok FJ, Grobbee DE, Geleijnse JM. Influence of weight reduction on blood pressure: a meta-analysis of randomized controlled trials. Hypertension. 2003;42(5):878-84.
131. Sacks FM, Svetkey LP, Vollmer WM, Appel LJ, Bray GA, Harsha D, et al. Effects on blood pressure of reduced dietary sodium and the Dietary Approaches to Stop Hypertension (DASH) diet. New England journal of medicine. 2001;344(1):3-10.
132. Whelton SP, Chin A, Xin X, He J. Effect of aerobic exercise on blood pressure: a meta-analysis of randomized, controlled trials. Annals of internal medicine. 2002;136(7):493-503.
133. Orlandi M, Graziani F, D'Aiuto F. Periodontal therapy and cardiovascular risk. Periodontology 2000. 2020;83(1):107-24.
134. Ridker PM, MacFadyen JG, Everett BM, Libby P, Thuren T, Glynn RJ, et al. Relationship of C-reactive protein reduction to cardiovascular event reduction following treatment with canakinumab: a secondary analysis from the CANTOS randomised controlled trial. Lancet. 2018;391(10118):319-28.
135. Tardif J-C, Kouz S, Waters DD, Bertrand OF, Diaz R, Maggioni AP, et al. Efficacy and safety of low-dose colchicine after myocardial infarction. New England journal of medicine. 2019;381(26):2497-505.
136. Ridker PM, Everett BM, Pradhan A, MacFadyen JG, Solomon DH, Zaharris E, et al. Low-dose methotrexate for the prevention of atherosclerotic events. New England Journal of Medicine. 2019;380(8):752-62.
137. Cuspidi C, Tadic M, Grassi G, Mancia G. Treatment of hypertension: The ESH/ESC guidelines recommendations. Pharmacological research. 2018;128:315-21.

138. Hill AB. The environment and disease: association or causation? : Sage Publications; 1965.
139. Davey Smith G, Hemani G. Mendelian randomization: genetic anchors for causal inference in epidemiological studies. Human molecular genetics. 2014;23(R1):R89-R98.
140. Czesnikiewicz-Guzik M, Osmenda G, Siedlinski M, Nosalski R, Pelka P, Nowakowski D, et al. Causal association between periodontitis and hypertension: evidence from Mendelian randomization and a randomized controlled trial of non-surgical periodontal therapy. European heart journal. 2019;40(42):3459-70.
141. Nibali L, Di Iorio A, Tu YK, Vieira AR. Host genetics role in the pathogenesis of periodontal disease and caries. J Clin Periodontol. 2017;44 Suppl 18:S52-s78.
142. Epstein SE, Zhou YF, Zhu J. Infection and atherosclerosis: emerging mechanistic paradigms. Circulation. 1999;100(4):e20-e8.
143. Libby P, Loscalzo J, Ridker PM, Farkouh ME, Hsue PY, Fuster V, et al. Inflammation, immunity, and infection in atherothrombosis: JACC review topic of the week. Journal of the American College of Cardiology. 2018;72(17):2071-81.
144. Aoyama N, Suzuki JI, Kumagai H, Ikeda Y, Akazawa H, Komuro I, et al. Specific periodontopathic bacterial infection affects hypertension in male cardiovascular disease patients. Heart and Vessels. 2017:1-7.
145. Fine N, Chadwick J, Sun C, Parbhakar K, Khoury N, Barbour A, et al. Periodontal Inflammation Primes the Systemic Innate Immune Response. Journal of Dental Research. 2020:0022034520963710.
146. Czesnikiewicz-Guzik M, Nosalski R, Mikolajczyk TP, Vidler F, Dohnal T, Dembowska E, et al. Th1-type immune responses to Porphyromonas gingivalis antigens exacerbate angiotensin II-dependent hypertension and vascular dysfunction. British journal of pharmacology. 2019;176(12):1922-31.
147. Ishai A, Osborne MT, El Kholy K, Takx RA, Ali A, Yuan N, et al. Periodontal disease associates with arterial inflammation via potentiation of a hematopoietic-arterial axis. JACC: Cardiovascular Imaging. 2019;12(11 Part 1):2271-3.
148. Postadzhiyan AS, Tzontcheva AV, Kehayov I, Finkov B. Circulating soluble adhesion molecules ICAM-1 and VCAM-1 and their association with clinical outcome, troponin T and C-reactive protein in patients with acute coronary syndromes. Clinical biochemistry. 2008;41(3):126-33.
149. Miyajima S-i, Naruse K, Kobayashi Y, Nakamura N, Nishikawa T, Adachi K, et al. Periodontitis-activated monocytes/macrophages cause aortic inflammation. Scientific reports. 2014;4(1):1-9.
150. Gilgun-Sherki Y, Melamed E, Offen D. Oxidative stress induced-neurodegenerative diseases: the need for antioxidants that penetrate the blood brain barrier. Neuropharmacology. 2001;40(8):959-75.
151. Small HY, Migliarino S, Czesnikiewicz-Guzik M, Guzik TJ. Hypertension: focus on autoimmunity and oxidative stress. Free Radical Biology and Medicine. 2018;125:104-15.
152. Araujo M, Wilcox CS. Oxidative stress in hypertension: role of the kidney. Antioxidants & redox signaling. 2014;20(1):74-101.

153. Chapple IL, Matthews JB. The role of reactive oxygen and antioxidant species in periodontal tissue destruction. Periodontology 2000. 2007;43(1):160-232.
154. Graves D. The potential role of chemokines and inflammatory cytokines in periodontal disease progression. Clinical Infectious Diseases. 1999;28(3):482-90.
155. Brito LC, DalBo S, Striechen TM, Farias JM, Olchanheski Jr LR, Mendes RT, et al. Experimental periodontitis promotes transient vascular inflammation and endothelial dysfunction. Archives of oral biology. 2013;58(9):1187-98.
156. da Silva JC, Muniz FWMG, Oballe HJR, Andrades M, Rösing CK, Cavagni J. The effect of periodontal therapy on oxidative stress biomarkers: A systematic review. Journal of clinical periodontology. 2018;45(10):1222-37.
157. Hirst DG, Robson T. Nitric oxide physiology and pathology. Nitric Oxide. 2011:1-13.
158. Godo S, Sawada A, Saito H, Ikeda S, Enkhjargal B, Suzuki K, et al. Disruption of physiological balance between nitric oxide and endothelium-dependent hyperpolarization impairs cardiovascular homeostasis in mice. Arteriosclerosis, thrombosis, and vascular biology. 2016;36(1):97-107.
159. Galley JC, Straub AC. Redox control of vascular function. Arteriosclerosis, thrombosis, and vascular biology. 2017;37(12):e178-e84.
160. Napoli C, Ignarro LJ. Nitric oxide and pathogenic mechanisms involved in the development of vascular diseases. Archives of pharmacal research. 2009;32(8):1103-8.
161. Bryan NS, Tribble G, Angelov N. Oral microbiome and nitric oxide: the missing link in the management of blood pressure. Current hypertension reports. 2017;19(4):1-8.
162. Koch CD, Gladwin MT, Freeman BA, Lundberg JO, Weitzberg E, Morris A. Enterosalivary nitrate metabolism and the microbiome: Intersection of microbial metabolism, nitric oxide and diet in cardiac and pulmonary vascular health. Free Radical Biology and Medicine. 2017;105:48-67.
163. Bescos R, Ashworth A, Cutler C, Brookes ZL, Belfield L, Rodiles A, et al. Effects of Chlorhexidine mouthwash on the oral microbiome. Scientific reports. 2020;10(1):1-8.
164. Sánchez GA, Miozza VA, Delgado A, Busch L. Total salivary nitrates and nitrites in oral health and periodontal disease. Nitric Oxide. 2014;36:31-5.
165. Parwani S, Chitnis P, Parwani R. Salivary nitric oxide levels in inflammatory periodontal disease–A case-control and interventional study. International journal of dental hygiene. 2012;10(1):67-73.
166. Meschiari CA, Zuardi LR, Gomes VA, de Almeida GRC, Novaes Jr AB, Gerlach RF, et al. Salivary, blood and plasma nitrite concentrations in periodontal patients and healthy individuals before and after periodontal treatment. Clinica Chimica Acta. 2015;444:293-6.
167. Arimatsu K, Yamada H, Miyazawa H, Minagawa T, Nakajima M, Ryder MI, et al. Oral pathobiont induces systemic inflammation and metabolic changes associated with alteration of gut microbiota. Scientific reports. 2014;4:4828.
168. Sato K, Takahashi N, Kato T, Matsuda Y, Yokoji M, Yamada M, et al. Aggravation of collagen-induced arthritis by orally administered Porphyromonas gingivalis through modulation of the gut microbiota and gut immune system. Scientific reports. 2017;7(1):1-13.

169. Nakajima M, Arimatsu K, Kato T, Matsuda Y, Minagawa T, Takahashi N, et al. Oral administration of P. gingivalis induces dysbiosis of gut microbiota and impaired barrier function leading to dissemination of enterobacteria to the liver. PloS one. 2015;10(7):e0134234.
170. Blasco-Baque V, Garidou L, Pomié C, Escoula Q, Loubieres P, Le Gall-David S, et al. Periodontitis induced by Porphyromonas gingivalis drives periodontal microbiota dysbiosis and insulin resistance via an impaired adaptive immune response. Gut. 2017;66(5):872-85.
171. Castelli WA, Diaz-Perez R, Nasjleti CE, Caffesse RG. Effect of renovascular hypertension on the morphology of oral blood vessels. Oral Surgery, Oral Medicine, Oral Pathology. 1978;46(4):576-82.
172. Bastos MF, Brilhante FV, Gonçalves TED, Pires AG, Napimoga MH, Marques MR, et al. Hypertension may affect tooth-supporting alveolar bone quality: a study in rats. Journal of periodontology. 2010;81(7):1075-83.
173. Leite CL, Redins CA, Vasquez EC, Meyrelles SS. Experimental-induced periodontitis is exacerbated in spontaneously hypertensive rats. Clinical and Experimental Hypertension. 2005;27(6):523-31.
174. Bonato C, do-Amaral C, Belini L, Salzedas LMP, Oliveira SHPd. Hypertension favors the inflammatory process in rats with experimentally induced periodontitis. Journal of periodontal research. 2012;47(6):783-92.
175. Santos CF, Akashi A, Dionísio T, Sipert C, Didier D, Greene A, et al. Characterization of a local renin-angiotensin system in rat gingival tissue. Journal of periodontology. 2009;80(1):130-9.
176. Santos CF, Morandini AC, Dionísio TJ, Faria FA, Lima MC, Figueiredo CM, et al. Functional local renin-angiotensin system in human and rat periodontal tissue. PloS one. 2015;10(8):e0134601.
177. Grimes DA, Schulz KF. Bias and causal associations in observational research. The lancet. 2002;359(9302):248-52.
178. Zinman E. Dental and legal considerations in periodontal therapy. Periodontology 2000. 2001;25(1):114-30.
179. Zhu L, Petersen PE, Wang H-Y, Bian J-Y, Zhang B-X. Oral health knowledge, attitudes and behaviour of adults in China. International dental journal. 2005;55(4):231-41.
180. Nasir N, Ali S, Ullah U. Extent of awareness regarding systemic effects of periodontal disease among medical interns. Ann Pak Inst Med Sci. 2013;9(4):188-90.
181. Weidlich P, Cimões R, Pannuti CM, Oppermann RV. Association between periodontal diseases and systemic diseases. Brazilian oral research. 2008;22:32-43.
182. Montero E, Herrera D, Sanz M, Dhir S, Van Dyke T, Sima C. Development and validation of a predictive model for periodontitis using NHANES 2011–2012 data. Journal of clinical periodontology. 2019;46(4):420-9.
183. Preshaw PM. Detection and diagnosis of periodontal conditions amenable to prevention. BMC oral health. 2015;15(1):1-11.
184. Botelho J, Proenca L, Leira Y, Chambrone L, Mendes JJ, Machado V. Economic Burden of Periodontal Disease in Europe and the United States of America-An updated forecast. medRxiv. 2021.

185. Smits KP, Listl S, Plachokova AS, Van der Galien O, Kalmus O. Effect of periodontal treatment on diabetes-related healthcare costs: a retrospective study. BMJ Open Diabetes Research and Care. 2020;8(1):e001666.
186. Choi SE, Sima C, Pandya A. Impact of treating oral disease on preventing vascular diseases: a model-based cost-effectiveness analysis of periodontal treatment among patients with type 2 diabetes. Diabetes care. 2020;43(3):563-71.
187. Thomas O, Shipman K, Day K, Thomas M, Martin U, Dasgupta I. Prevalence and determinants of white coat effect in a large UK hypertension clinic population. Journal of human hypertension. 2016;30(6):386-91.
188. Pappaccogli M, Enri LR, Perlo E, Di Monaco S, Pignata I, Baratta F, et al. Assessment of a non-physician screening program for hypertension and cardiovascular risk in community pharmacies. Nutrition, Metabolism and Cardiovascular Diseases. 2019;29(12):1316-22.
189. Stergiou GS, Parati G, McManus RJ, Head GA, Myers MG, Whelton PK. Guidelines for blood pressure measurement: development over 30 years. The Journal of Clinical Hypertension. 2018;20(7):1089-91.
190. Paini A, Bertacchini F, Stassaldi D, Aggiusti C, Maruelli G, Arnoldi C, et al. Unattended versus attended blood pressure measurement: mean values and determinants of the difference. International journal of cardiology. 2019;274:305-10.
191. Levine R, Stillman-Lowe CR. The scientific basis of oral health education: Springer; 2019.

Supplementary data

9. SUPLEMENTARY DATA

9.1. THE 1st ORIGINAL PUBLICATION

9.2. THE 2nd ORIGINAL PUBLICATION

9.3. THE 3rd ORIGINAL PUBLICATION

9.4. LECTURE FOR FEDERAL UNIVERSITY OF PARANÁ (BRASIL)

9.5. LECTURE FOR SEPA: ALIANZA POR LA SALUD ORAL Y GENERAL

The 1st original publication

ESC European Society of Cardiology
Cardiovascular Research
doi:10.1093/cvr/cvz201

REVIEW

Periodontitis is associated with hypertension: a systematic review and meta-analysis

Eva Muñoz Aguilera [1,2], Jean Suvan[1], Jacopo Buti [1], Marta Czesnikiewicz-Guzik[3,4,5,6], Aline Barbosa Ribeiro [3,4,7], Marco Orlandi[1], Tomasz J. Guzik[3,4,5,6], Aroon D. Hingorani[8], Jose Nart [2], and Francesco D'Aiuto [1]*

[1]Periodontology Unit, University College London Eastman Dental Institute, London WC1X 8LD, UK; [2]Department of Periodontology, Faculty of Dentistry, Universitat Internacional de Catalonia, Sant Cugat del Vallès, Barcelona 08195, Spain; [3]Department of Periodontology and Oral Sciences Research Group, University of Glasgow Dental School, Glasgow G12 8QQ, UK; [4]Institute of Cardiovascular and Medical Sciences, University of Glasgow, Glasgow G12 8QQ, UK; [5]Department of Experimental Dentistry and Dental Prophylaxis, Jagiellonian University, Krakow, Poland; [6]Department of Internal and Agricultural Medicine, Jagiellonian University, Krakow, Poland; [7]Department of Physiology, Ribeirao Preto Medical School, University of Sao Paulo, Sao Paulo, Brazil; and [8]Genetic Epidemiology, Institute of Cardiovascular Science, Faculty of Population Health Sciences, University College London, London WC1E 6BT, UK

Received 30 May 2019; revised 12 July 2019; editorial decision 26 July 2019; accepted 29 July 2019

This article was handled by Consulting Editor, Giuseppe Lembo.

Abstract

Recent evidence suggests a link between periodontitis (PD) and hypertension, but the nature of this association remains unclear. The overall aim of this review was to critically appraise the evidence linking these two common disorders. Systematic search was conducted for studies published up to December 2018. Prevalence of hypertension in patients with PD (moderate/severe groups) vs. those without PD (non-PD) was the primary outcome. Additional outcomes included adjusted mean difference in systolic (SBP) and diastolic (DBP) blood pressure (BP) levels in PD vs. non-PD, assessment of biomarkers in PD and hypertension, and BP changes after periodontal therapy. From 81 studies selected, 40 were included in quantitative meta-analyses. Diagnoses of moderate-severe PD [odds ratio (OR) = 1.22; 95% confidence interval (CI): 1.10–1.35] and severe PD (OR = 1.49; 95% CI: 1.09–2.05) were associated with hypertension. Prospective studies confirmed PD diagnosis increased likelihood of hypertension occurrence (OR = 1.68; 95% CI: 0.85–3.35). Patients with PD exhibited higher mean SBP [weighted mean difference (WMD) of 4.49 mmHg; 95% CI: 2.88–6.11] and DBP (2.03 mmHg; 95% CI: 1.25–2.81) when compared with non-PD. Lastly, only 5 out of 12 interventional studies confirmed a reduction in BP following periodontal therapy, ranging from 3 to 12.5 mmHg of SBP and from 0 to 10 mmHg of DBP. PD is associated with increased odds of hypertension (SORT C) and higher SBP/DBP levels. The evidence suggesting that PD therapy could reduce BP is inconclusive. Although additional research is warranted on this association, these results suggest that oral health assessment and management of PD could not only improve oral/overall health and quality of life but also be of relevance in the management of patients with hypertension.

Keyword Hypertension • Periodontitis • Blood pressure • Inflammation • Periodontal diseases • Oral health • Periodontal therapy

1. Introduction

Hypertension, defined as values ≥140 mmHg systolic blood pressure (SBP) and/or ≥90 mmHg diastolic blood pressure (DBP), is the most prevalent of all cardiovascular diseases (CVDs).[1] Almost 45% of the worldwide population is affected and the estimate increases steeping with age.[2] The incidence of adverse cardiovascular (CV) events such as stroke, myocardial infarction, sudden death, heart failure, and peripheral artery disease as well as of end-stage renal disease is strongly associated with hypertension.[3,4] According to the World Health Organization (WHO) report in 2014, hypertension accounts for 51% of deaths from stroke and 45% of overall CV mortality and this is true at all ages and in all ethnic groups.[2] Blood pressure values are an important predictor of cardiovascular risk.[5,6] Despite available treatments, essential hypertension remains poorly controlled with high rates of no treatment and under-treatment.[7] Hence, it is still one of the major modifiable risk factor for CVDs that requires urgent management.[8] Hypertension is a complex multifactorial disease with no simple mechanism entirely explaining the blood pressure rise.[9] Endothelial dysfunction (as manifested by changes in endothelin and nitric oxide), oxidative stress, and inflammation are

* Corresponding author. Tel: +44 203 456 1108; fax: +44 203 456 1137, Email: f.daiuto@ucl.ac.uk

implicated in the development of hypertension. Despite a prominent role of the immune system being observed in experimental models[10] and clinical studies[11] studying the onset of hypertension, the exact mechanisms initiating these responses remain unclear.[12]

Periodontitis is a chronic multifactorial inflammatory disease caused by a dysbiotic microflora and resulting in progressive destruction of the dental surrounding tissues and leading to tooth loss. It is associated with masticatory dysfunction and negative impact on the patient's quality of life.[13] It is estimated that periodontitis affects over 50% of the worldwide population and its severe form is considered the 6th most prevalent disease of humankind.[14,15] Periodontitis is a major public health problem that considerably increases morbidity and costs of oral healthcare.[16,17] There is consistent observational evidence that periodontitis is associated with an increased risk for future CVDs independent of traditional risk factors such as smoking and obesity.[17,18] The interplay between the bacterial burden and host response is the most plausible biological mechanism linking periodontitis to a number of chronic systemic diseases, such as diabetes mellitus, CVDs, and neurological diseases such as Alzheimer.[17,19,20] An ulcerated epithelial lining of the gingival pocket, subsequent to a local inflammatory response to the dental biofilm could amount to a sizeable area in patients with generalized periodontitis.[21] These patients often present with systemic inflammation and endothelial dysfunction,[22] which improves following successful periodontal treatment.[23]

Several studies appear to support a relationship between severe periodontitis and hypertension.[24–27] Limited evidence also suggests that successful periodontal treatment could improve arterial blood pressure.[28,29] However, little is still known about the direction and nature of the association between these two conditions. The overall aim was to conduct a robust critical appraisal of the evidence on the relationship between periodontitis and hypertension. Specific research questions were designed based on the following PECO outline: *Population*: Individuals >16 years old; *Exposure*: Presence of periodontitis with/without treatment; *Comparison*: Individuals with no periodontitis; *Outcome(s)*: Any measure of prevalence and/or levels of hypertension and/or changes in blood pressure following periodontal therapy. In this analysis we addressed several key questions:

- Are patients with periodontitis more likely to have hypertension (compared to those without periodontitis)?
- Is the degree of hypertension influenced by the severity and/or extent of periodontitis (linear association)?
- Is the mean SBP/DBP higher in patients with periodontitis vs. those without periodontitis?
- Does periodontal therapy modify the levels of blood pressure?

2. Methods

The systematic review protocol was registered in PROSPERO on 28/11/2017 with ID: CRD42017081455. A PRISMA statement is attached to follow the reporting of this systematic review (Supplementary material online, *Appendix S1*).

2.1 Primary and secondary outcomes

The primary outcome of this systematic review was odds ratio (OR)/relative risk (RR) and confidence interval (CI) for hypertension in individuals with periodontitis.

The secondary outcomes included prevalence of hypertension in patients with periodontitis vs. patients without periodontitis as well as prevalence of periodontitis in patients with or without hypertension; reports of mean SBP/DBP levels in periodontally healthy and diseased patients; systemic biomarkers associated with periodontitis and hypertension and changes in BP measurements following periodontal therapy.

2.2 Inclusion/exclusion criteria

To obtain an estimate of the association between periodontitis and hypertension, inclusion criteria were set to be broad and inclusive. Prospective and retrospective studies were included (randomized controlled trials, controlled clinical trials, cohort studies, case–control studies, and cross-sectional studies). Eligibility criteria included individuals from age 16 years onwards, with periodontitis (chronic and/or aggressive forms) considered as the exposure. Manuscripts including information related to primary and secondary outcomes were included.

Case report, case series and reviews, and animal studies were excluded. Individuals under 16 years old and pregnant women were also excluded. Lastly, studies that did not have any reports of the primary or secondary outcomes were disqualified.

2.3 Search methods for identification of studies

Five electronic databases were searched up to 10 December 2018 with no year restrictions [Cochrane Central Register of Controlled Trials (CENTRAL), MEDLINE (OVID), EMBASE, Web of Science and Latin American & Caribbean Health Sciences Literature (LILACS)]. The search included no language restrictions and attempts were made to translate non-English language manuscripts (if this was not possible then the relevant evidence was excluded). In addition, SIGLE database was examined for relevant unpublished trials. Performed detailed search strategies deemed appropriate for each database using a combination of controlled vocabulary (MeSH terms and free text terms). All terms are available in the Supplementary material online, *Appendix S2 Table*).

MeSH terms in all trees and subheadings: 'periodontal diseases', 'periodontics', 'hypertension', 'blood pressure'.

Keywords: 'periodont$', 'gingiv$', '((blood or bleed$) adj4 prob$)', '(ging$adj disease)', 'hypertens$', '((elevat$or high$or rais$) adj3 (diastolic or systolic or arterial or blood) adj pressure)', 'bloodpressure'.

Hand searching of bibliographies of papers and review articles retrieved articles not found through other search methods.

2.4 Data management

The eligibility assessment of titles and abstracts (when available) of all reports identified were independently screened by two reviewers based on inclusion/exclusion criteria (E.M.A. and J.S.). If agreement could not be reached, the study was moved to the next stage and inclusion was based on full text screening. Full reports were obtained and assessed independently and in duplicate (E.M.A. and J.S.) for studies seeming to meet the inclusion criteria or for which insufficient information in the title and abstract precluded to make a clear decision. Disagreements were resolved by discussion and if necessary, a third reviewer was consulted (F.D.). When authors were not reporting on an effect estimate they were contacted to request additional information. Excel sheets were created to document information regarding decision for included and excluded articles. Kappa statistic was used to assess the agreement between the reviewers based on full text screening.

The main categories of data grouped according to study design and reported in evidence tables were study characteristics data; population; exposure (case definition for periodontitis); outcome (case definition for hypertension); effect (OR/RR with CI); and publication conclusions.

Regarding the exposure, multiple case definitions for periodontitis were found. A lack of consistent case definitions contributed to the difficultly in assessment and interpretation of the data retrieved. In order to collate studies looking at similar definitions, results were therefore grouped using two case definition thresholds: confident and non-confident case definition of periodontitis based on the following criteria (adapted from a previously reported definition).[30]

2.4.1 Confident case definition of periodontitis

The following criteria were considered as a confident case definition for periodontitis: generalized chronic periodontitis (at least 30% sites with CAL ≥ 4 mm)[31]; at least two sites on different teeth with clinical attachment level (CAL) 6 mm and at least one site with probing pocket depth (PPD) 4 mm (CDC/AAP periodontitis definition)[32]; presence of proximal attachment loss of >3 mm in two or more non-adjacent teeth (sensitive definition) or presence of proximal attachment loss of >5 mm in >30% of teeth present[33]; at least five sites with CAL ≥ 6 mm.[34]

2.4.2 Non-confident case definition of periodontitis

For non-confident case definition the following reported criteria were considered: community periodontal index (CPI) score 3/4 in at least one quadrant; 'Alveolar bone loss' without other measurements of PPD/CAL; unclear diagnostic criteria for periodontitis.

2.4.3 Definition of hypertension

Regarding the outcome, hypertension was defined as SBP ≥140 mmHg/DBP ≥90 mmHg or the use of anti-hypertensive medications.[1,2] However, reports of BP levels and other cases definitions were also documented for, such as self-reported hypertension and other thresholds (high normal/pre-hypertension).

2.5 Assessment of bias individual studies

Quality assessment of all included studies was undertaken independently and in duplicate by two reviewers as part of the data extraction process. For bias assessment of randomized controlled trials, non-randomized studies of interventions, and observational studies we used the revised Cochrane tool (ROB-2.0 tool),[35] the ROBINS-I tool,[36] and the Newcastle–Ottawa (NOS) tool,[37] respectively.

2.6 Data synthesis

Descriptive statistics were performed to summarize the evidence retrieved and to determine the quantity of data, checking further for study variations in terms of study characteristics and results. This assisted in confirming the suitability of further synthesis methods.

Meta-analysis A was conducted and referred to the following primary outcome: ORs for hypertension among people with or without a diagnosis of periodontitis. The ORs with adjustment for the confounding variables (i.e. age, gender, smoking, socioeconomic status, systemic disease, medication, body mass index, etc.) was chosen with hypertension as the dependent variable and periodontitis as the independent variable. Pooled estimates of OR and corresponding 95% confidence intervals were calculated for dichotomous data. In presence of significant heterogeneity ($P < 0.1$), the pooled estimates of effects were calculated using random effects models rather than fixed effects models. Meta-analysis B referred to the secondary outcome (mean SBP/DBP). The pooled mean SBP/DBP difference and 95% confidence intervals were estimated for continuous data. RevMan® 5.3 and JMP® 13.0.0 were used for all the statistical analyses.

To evaluate whether the methodological quality of the included studies influenced the direction or the magnitude of the results, we performed a separate sensitivity analysis by study design and either disease severity or case definition (*Figures 2* and *3C* and Supplementary material online, *Appendix S5*).

2.7 Publication bias

Possible publication bias was assessed for studies included in the different meta-analyses A and B using the methods described by Begg *et al.* and Egger *et al.*[38,39]

2.8 Heterogeneity

The significance of any discrepancies in the estimates from different trials was assessed by means of Cochran's test for heterogeneity and the I^2 statistic. As alluded above, sensitivity analyses were also planned to explore, quantify, and control for sources of heterogeneity between studies.

2.9 Strength of recommendation

The quality and strength of the evidence was assessed with The SORT (Strength of Recommendation Taxonomy). The authors discussed the outcomes of the systematic review, pertinent sources of evidence, clinical recommendations, and future areas requiring research.[40]

3. Results

The electronic search from combination of all databases identified 5574 potentially relevant articles after removal of duplicates, resulting in 182 publications eligible for full text screening. Eighty-one publications met inclusion criteria (*Figure 1*). The evidence tables created according to study design, included the main study characteristics (Supplementary material online, *Appendix S3*). The studies included in the systematic review have been conducted in 26 different countries from all continents involving a large variety of different populations. Reviewers (E.M.A. and J.S.) achieved an almost perfect agreement with 97.24%; Cohen's *k*: 0.94.

A variety of case definition of periodontitis was identified (as shown in evidence tables, Supplementary material online, *Appendix S3*). For hypertension diagnosis, the studies generally reported more uniformed criteria based on levels of SBP ≥140mmHg and/or DBP ≥90 mmHg or use of anti-hypertensive medication.[41] Nevertheless, some studies reported lower cut offs for hypertension (i.e. SBP ≥130mmHg and/or DBP ≥85 mmHg)[42] or hypertension prevalence was based on medical records, self-report, or national classification codes for disease. Similarly, different methods for measuring blood pressure were described in the studies included (Supplementary material online, *Appendix S3*). For additional or missing data, of all the authors contacted, only three provided additional information regarding the direction of the association and/or mean SBP/DBP following periodontal therapy.[43–45]

Study quality for observational studies as assessed by the Newcastle–Ottawa scale varied across the studies, ranging from a score of 3/9 to 9/9 (Supplementary material online, *Appendix S4*). The assessment revealed several potential sources of bias including the adequacy of case definition for cases and controls, the representativeness of the cases, no appropriate description of the sample size calculation, lack of adjustment for potential confounders or inappropriate statistical test. The assessment of randomized controlled trials with the Rob 2.0 tool revealed a low (five studies) to high (two studies) risk of bias for the studies included (Supplementary material online, *Appendix S4*). The main reasons for high risk of bias in randomized controlled trials arose from the randomization

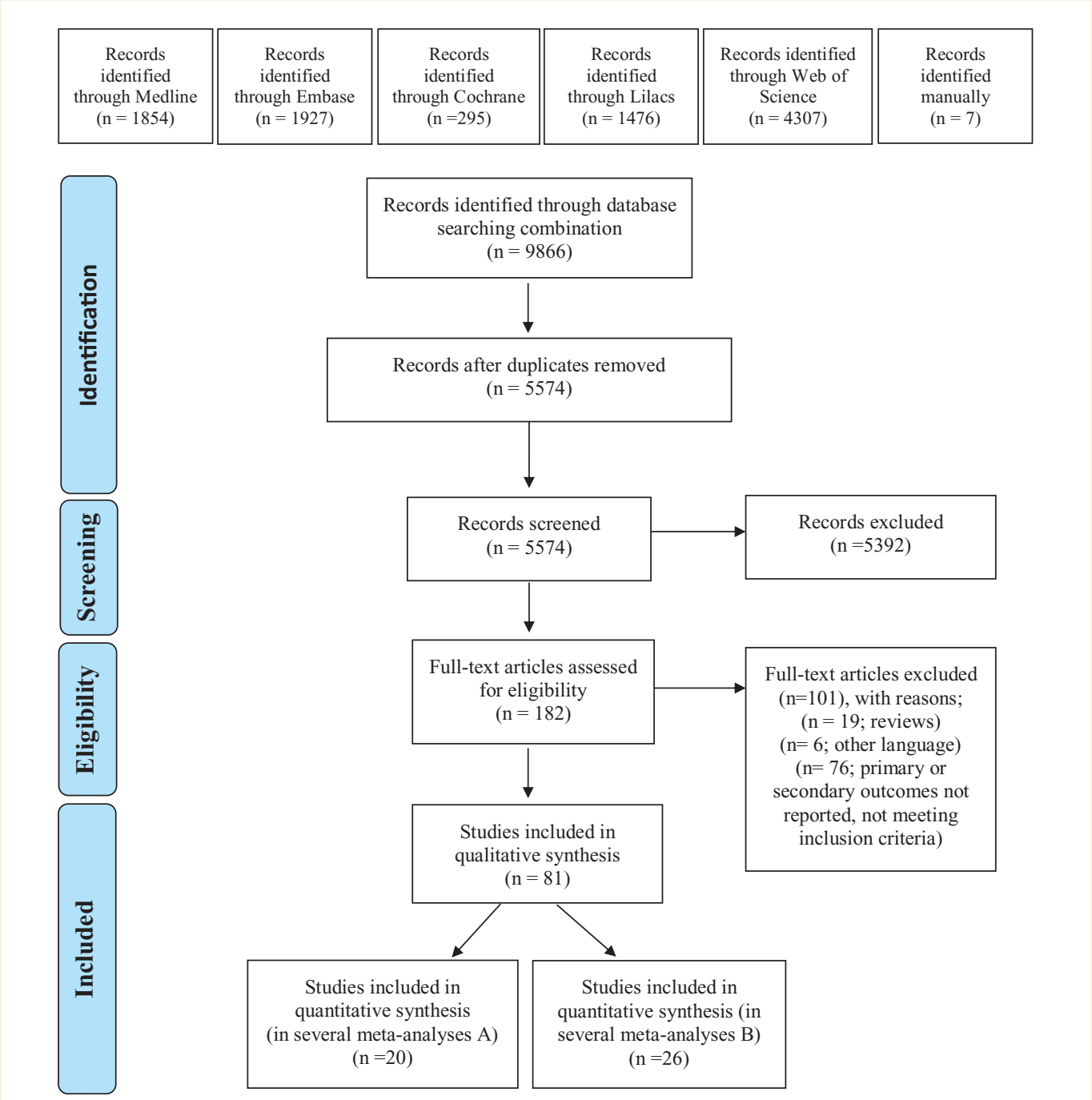


Figure 1 PRISMA flowchart. Flowchart of the study selection process. A systematic review yielded 9866 reports. After removal of duplicates and the application of inclusion and exclusion criteria, 46 studies were included in two different meta-analyses. PubMed, Embase, Cochrane, Lilacs, Web of Science, and manual search strategies are illustrated in Supplementary material online, *Appendices S1 and S2*.

process, blinding of participants and personnel. Study quality for non-randomized trials revealed moderate and serious risk of bias for the two studies assessed with the Robinson I tool (Supplementary material online, *Appendix S4*).

3.1 Primary outcome

Twenty studies included in five meta-analyses (A) of cohort, cross-sectional and case–control studies (*Figures 2* and *3C* and Supplementary material online, *Appendix S5*) compared the odds of having hypertension if an individual had periodontitis vs. periodontally healthy individuals using a periodontal case definition as the exposure measure.

Statistically significant heterogeneity was confirmed with a τ^2 test (ranging from 0.32 to 0.03), χ^2 test ranging from (ranging from <0.00001 to 0.008), and I2 test (ranging from 63% to 92%) for the different analyses completed. Due to this level of heterogeneity observed in the studies, random effect meta-analysis was performed.

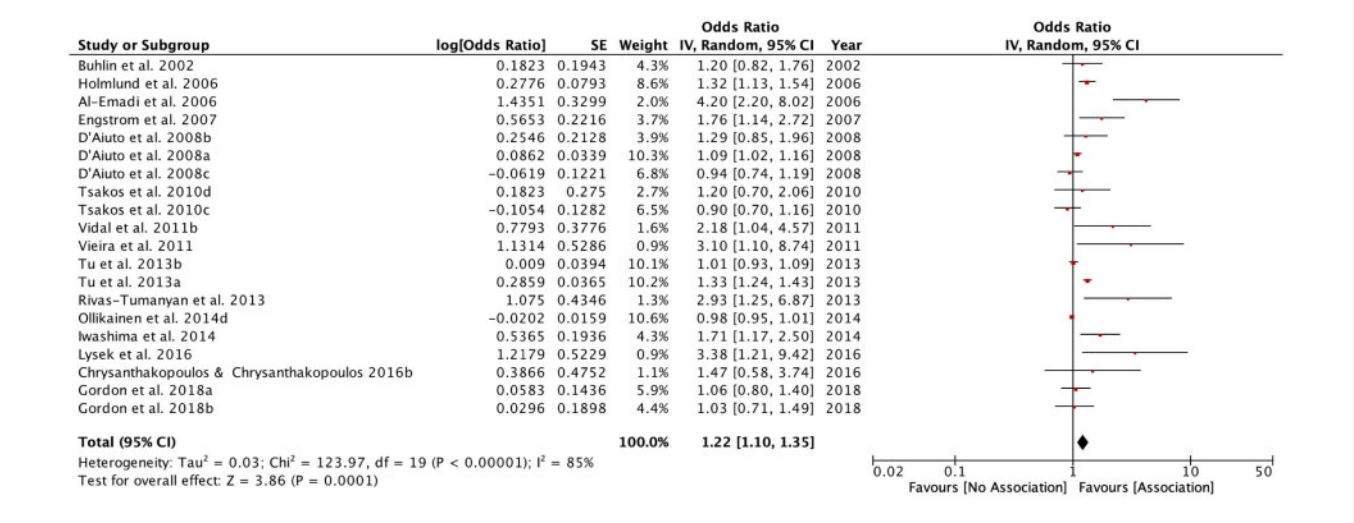


Figure 2 Association between periodontitis (moderate to severe combined diagnosis) and hypertension (cross-sectional and case–control studies). Summary Forrest plot for odds ratio of hypertension in relation to periodontitis diagnosis in cross-sectional and case–control studies (moderate to severe combined diagnosis). The random effects model was used and the relative size of the data markers indicates the weight of the sample size from each study. CI, confidence interval; IV, inverse variance; SE, standard error.

Odds ratios ranged from 0.90 to 4.20 for all studies, depending on case definition applied, severity of periodontitis and adjustment of the models. Precision of the estimates in the studies varied considerably as appreciated in the varying span of the confidence intervals. Two studies[24,46] reported ORs for moderate to severe periodontitis separately and one study[47] reported OR for men and women also separately, therefore these different ORs were included independently.

The analysis of three cohort studies predicted the occurrence of hypertension (OR = 1.68; 95% CI: 0.85–3.35), but this was not statistically significant ($P = 0.14$) (Supplementary material online, *Appendix S5*). Three studies were excluded from this meta-analysis due to one of them reported RR[48] and the other two appeared to be duplicated data.[45,49] Diagnosis of moderate-severe periodontitis in 15 cross-sectional and case–control studies was associated with higher odds of hypertension (1.22, 95% CI: 1.10–1.35), which was statistically significant ($P = 0.0001$) (*Figure 2*). A meta-analysis of eight cross-sectional and case–control studies confirmed that patients with severe periodontitis had increased odds (1.49, 95% CI: 1.09–2.50; $P = 0.01$) of diagnosis of hypertension (*Figure 3A*). Additionally, meta-analyses of studies according to confident vs. non-confident case definitions of periodontitis were performed. Seven studies with confident definition of periodontitis confirmed higher odds of hypertension (1.53, 95% CI: 1.11–2.10; $P = 0.009$) compared to a meta-analysis of eight studies with a non-confident definition of periodontitis (1.33, 95% CI: 1.14–1.55; $P = 0.003$) (*Figure 3B* and *C*).

3.2 Secondary outcomes

3.2.1 Prevalence

Thirty studies reported prevalence of hypertension in patients with periodontitis vs. patients without periodontitis or gingivitis (Supplementary material online, *Appendix S6*). Twenty-five of these studies showed a higher prevalence of hypertension in patients with a diagnosis of periodontitis (range = 7–77%) vs. those without periodontitis (range = 4–70%) and one study only confirmed higher prevalence in men.[50] These findings were not confirmed in four studies.[51–54] In addition, a consistent increased prevalence of periodontitis in patients with hypertension (range = 29–61%) vs. those without hypertension (range = 17–39%) was reported in all the seven publications that included this outcome (Supplementary material online, *Appendix S6*).

3.2.2 Mean blood pressure (observational evidence)

Thirty-one studies reported average mean SBP/DBP in patients with (range SBP = 113–172/DBP = 66–101 mmHg) and without periodontitis (range SBP = 109–143/DBP = 65–94 mmHg) (Supplementary material online, *Appendix S7*). The meta-analysis B, of mean SBP/DBP of 26 studies was performed resulting in statistically significant heterogeneity, confirmed with a Tau-squared test (ranging from 14.38 to 2.92), χ^2 test ranging from (<0.00001), and I2 test (ranging from 96 to 98%). Patients with periodontitis exhibited higher SBP [weighted mean difference (WMD) of 4.49 mmHg, 95% CI: 2.88–6.11; $P < 0.00001$] and DBP (WMD of 2.03 mmHg, 95% CI: 1.25–2.81; $P < 0.00001$) when compared with patients without periodontitis (*Figures 4* and *5*).

3.2.3 Systemic biomarkers

Three studies were included in the review as reporting changes in systemic biomarkers associated with hypertension and periodontitis.[55–57] One study analysed serum levels of neutrophilic enzymes in 95 patients.[55] They included a test group of patients with hypertension and periodontitis and two control groups: a healthy group (without periodontitis or hypertension) and a hypertensive group. The authors observed that circulating levels of matrix metalloproteinases (MMP)-8, MMP-9, myeloperoxidase and neutrophil elastase (NE) were increased in patients with hypertension and periodontitis but not in the controls. Another study examining the gingival crevicular fluid levels in patients with hypertension (21 patients) and without hypertension (26 patients) measuring levels of 8-isoprostane, interleukin (IL)-1B, monocyte chemoattractant protein (MCP)-1, tumour necrosis factor (TNF)α, C-reactive protein (CRP), and MMP-8.[56] They reported that independent of hypertension present or absent, an increased level of these biomarkers was

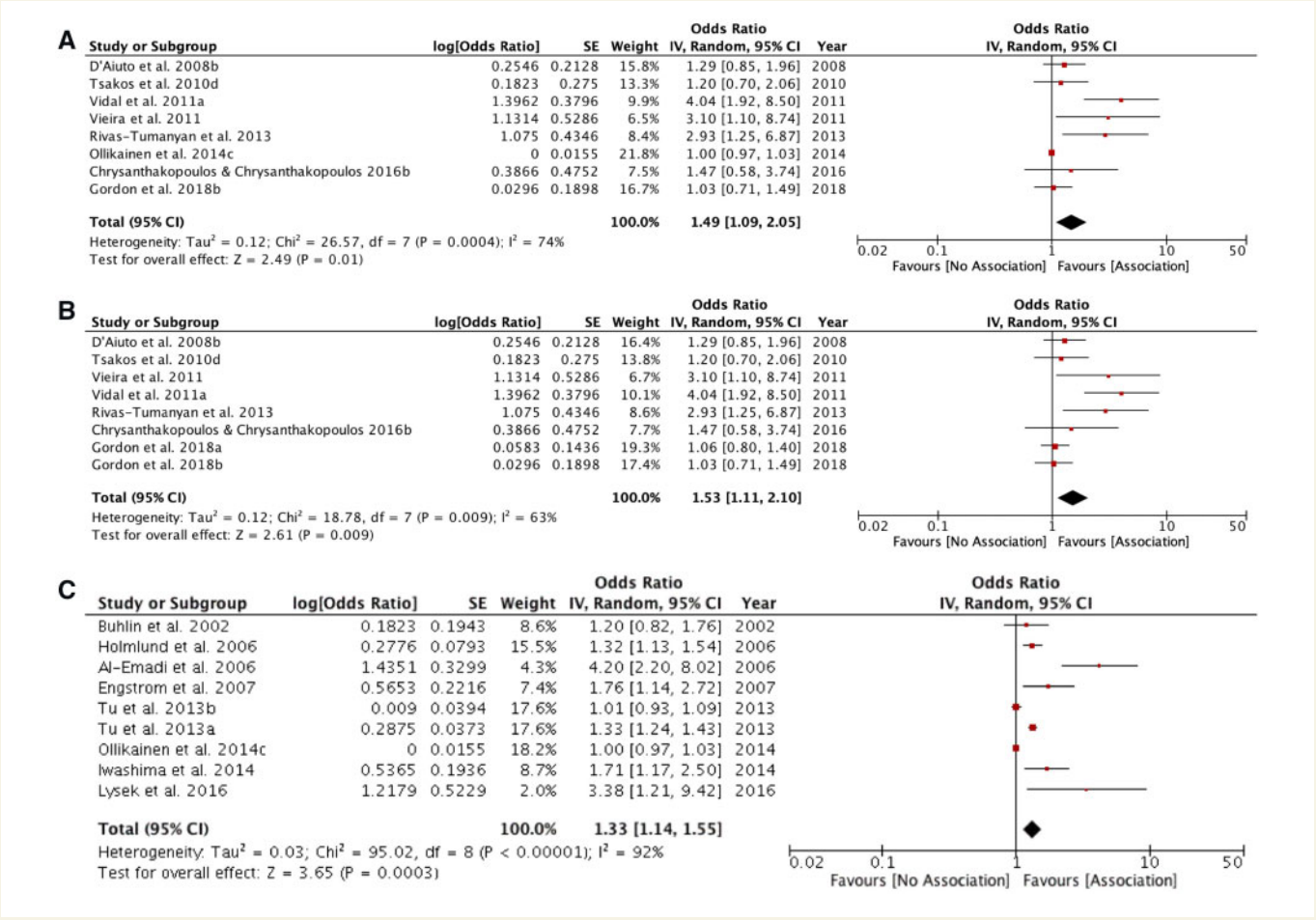


Figure 3 (*A–C*) Association between periodontitis (severe, confident, and non-confident diagnosis) and hypertension. Subgroup analysis Forrest plots for odds ratio of hypertension in relation to periodontitis status in cross-sectional and case–control studies. (*A*) Severe periodontitis only group adjusted. (*B*) Analysis adjusted for confident definition of periodontitis as described in methods. (*C*) Analysis adjusted for non-confident definition of periodontitis as described in methods. The random effects model was used and the relative size of the data markers indicates the weight of the sample size from each study. CI, confidence interval; IV, inverse variance; SE, standard error.

observed when patients had periodontal pockets. In addition, patients with hypertension presented with almost twice as much periodontal clinical attachment loss (CAL) as controls (Mean + SEM in HTN = 0.87 ± 0.13 vs. non-HTN = 0.49 ± 0.11). Albush *et al.*[57] assessed levels of vascular thrombotic markers in 40 hypertensive patients with periodontitis. Platelet count, fibrinogen, Von Willebrand factor antigen (vWF:Ag), and D-Dimer levels increased after 48 h of treatment (scaling of the teeth including subgingival root debridement for half of the patients and surgical periodontal therapy for the other 20) and decreased after 6 weeks ($P < 0.05$), with no significant differences between groups ($P > 0.05$). Acute increase in endothelial-activation markers including E-selectin, vWF, haemoglobin and haematocrit, D-dimer levels, and neutrophils counts was also reported 24 h following periodontal therapy in several publications.[22,44,58] Reductions in inflammatory biomarkers were observed in 11 interventional studies following periodontal therapy.[22,28,29,44,59–65]

3.2.4 Mean blood pressure (interventional evidence)

The search located 12 interventional clinical trials reporting the effect of periodontal therapy on blood pressure either as a primary[65] or secondary outcome (the remaining 11 studies) (see Supplementary material online, *Appendix S3* for a detailed description of the studies and treatment modalities). Eight studies were RCTs, three were non-RCT, and one was a pilot study. These studies comprised a varied sample of individuals, including people medically healthy in six studies[22,28,44,58,59,62] pre-hypertension,[65] refractory hypertension,[29] hypertension,[59] metabolic syndrome,[63] coronary artery disease,[60] and Type 2 diabetes.[64] Five of the 12 interventional studies included in the analysis confirmed a reduction in SBP following periodontal therapy (range = 3–12.5 mmHg), and an inconsistent reduction of DBP (range = 0–10 mmHg).[28,29,61,64,65] Six studies reported no changes in blood pressure measures following non-surgical and/or surgical periodontal therapy[44,58–60,62,63], however only two studies out of these six reported actual blood pressure values[59,60] and one author provided values upon request[44]. One study[22] reported an increase in blood pressure in the test group 1 day after periodontal therapy.

3.3 Publication bias

Study publication bias was examined using funnel plots for both meta-analyses A and B (Supplementary material online, *Appendix S8*). Egger's

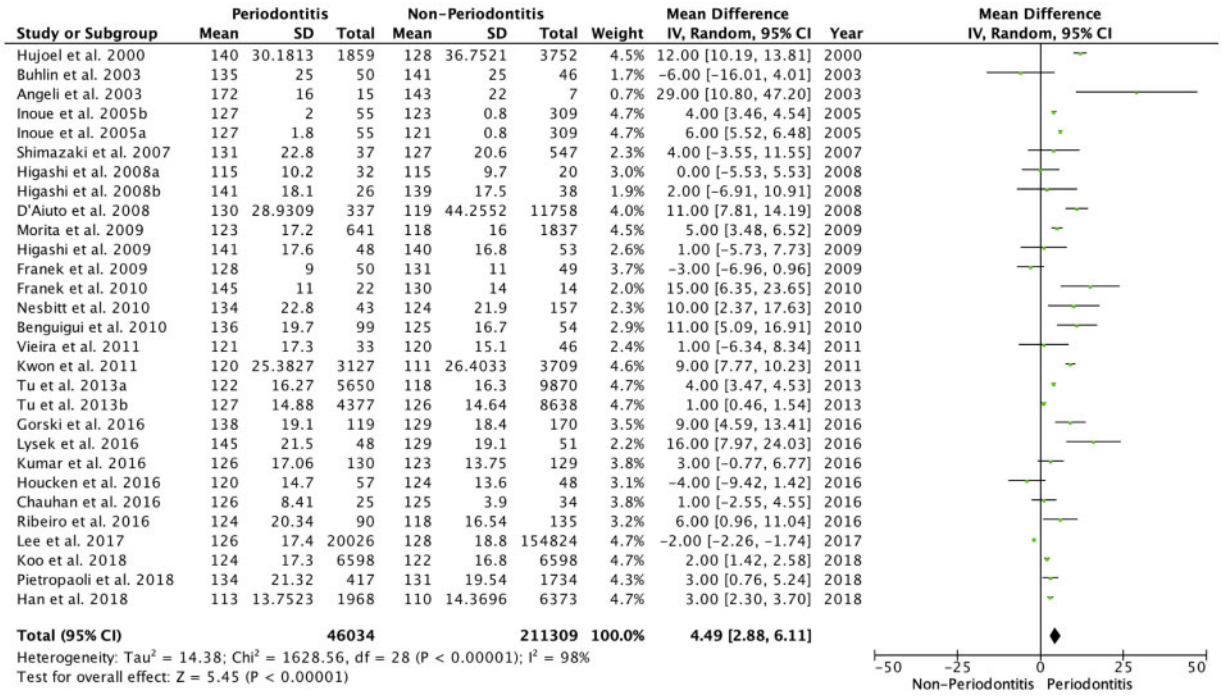


Figure 4 Periodontitis effect on systolic blood pressure (SBP). Summary Forrest plot for change in SBP in relation to periodontitis status in cross-sectional and case–control studies. The random effects model was used, weighted mean difference (WMD) reported and the relative size of the data markers indicates the weight of the sample size from each study. SE, standard error; IV, inverse variance; CI, confidence intervals.

test was only calculated for the meta-analyses A of moderate to severe periodontitis (Supplementary material online, *Appendix S8*). Visual assessment of the Funnel of moderate to severe periodontitis revealed studies were slightly skewed to the right, which was confirmed with the Egger's test showing a statistically significant difference ($P = 0.0054$); publication bias was therefore suspected in this analysis. Nevertheless, all the other funnel plots for meta-analyses A displayed symmetrical appearance. Similarly, visual assessment of the Funnel plots for mean SBP and DBP analysis revealed symmetrical appearance. Egger's test estimated for meta-analyses B revealed a not statistically significant result ($P = 0.5582$) for the mean SBP meta-analysis and a statistically significant difference ($P = 0.0224$) for the mean DBP meta-analysis. On this basis, publication bias was suspected in the mean DBP meta-analysis.

3.4 Reporting on strength of recommendation

The quality, quantity, and consistency of the evidence from observational and interventional studies on the relationship between periodontitis and hypertension were thoroughly assessed for a SORT recommendation. Accordingly, we conclude that diagnosis and treatment of periodontitis is positively associated with hypertension (SORT C).[40]

4. Discussion

The results of this systematic review support a positive association between periodontitis and hypertension. Based on the quantitative analyses of all studies included, patients with moderate to severe periodontitis have greater (20%) odds of having hypertension when compared to patients without periodontitis. In addition, a positive linear association was observed, confirming that the more severe periodontitis is, the higher the likelihood (49%) of having hypertension. This finding was further corroborated, when the studies with a confident case definition for periodontitis were analysed, confirming even greater odds (50%) of diagnosis of hypertension were found. The magnitude of association between periodontitis and hypertension reported in this review (OR 1.22–1.53) is in agreement with that recently reported.[27] In this recent review, however, Martin-Cabezas *et al.* included observational studies without specifying the exposure and outcome of the analysis. In the current systematic review, we also included three cohort studies[66–68] confirming a temporal association between periodontitis and incidence of hypertension although this was not statistically significant and we excluded a number of studies in this analysis in order to avoid bias due to suspected duplication of data.

This systematic review also confirmed an increased prevalence of periodontitis in patients with hypertension (as defined by SBP ≥140 and DBP ≥90 mmHg). Clinical and experimental evidence suggest that this direction of the association could be mediated through hypertension causing microcirculatory changes in of the gingival tissue leading to ischaemia, increased inflammation, and/or altered microbial composition of the dental biofilm.[25,69,70] This finding combined with the increased prevalence of hypertension in patients with periodontitis could be even more significant within the context of the new revised guidelines issued by the AHA for the definition of hypertension.[71] A reduced threshold of SBP/DBP for the case definition of hypertension was expressed (Stage 1 as SBP = 130–139/DBP = 80–89 mmHg, and Stage 2 agreeing to Stages 1

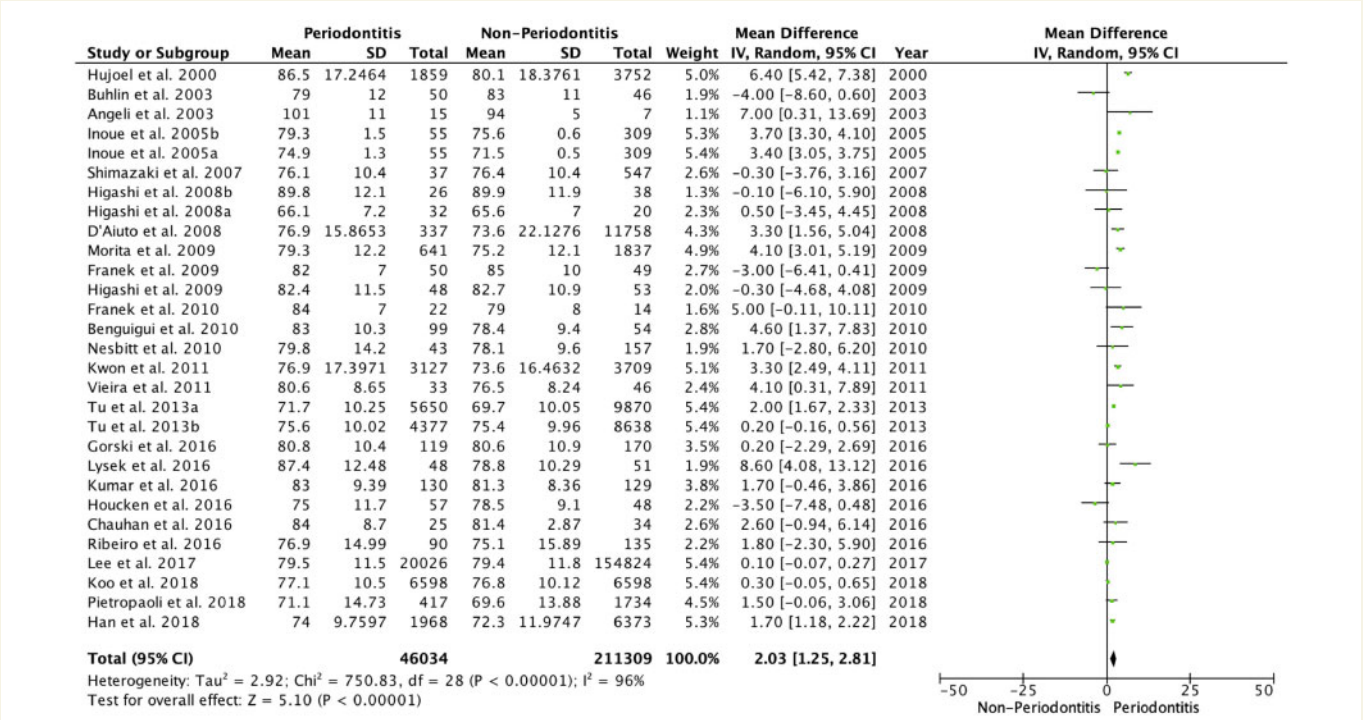


Figure 5 Periodontitis effect on diastolic blood pressure (DBP). Summary Forrest plot for change in DBP in relation to periodontitis status in cross-sectional and case–control studies. The random effects model was used, WMD reported and the relative size of the data markers indicates the weight of the sample size from each study. SE, standard error; IV, inverse variance; CI, confidence intervals.

and 2 in the JNC 7 report; i.e. SBP ≥ 140/DBP ≥ 90 mmHg), which has been reported in a recent cross-sectional study to double the prevalence estimates of hypertension in countries like China and USA.[72] This could result in even greater odds of hypertension in patients with periodontitis and vice versa. Future research should consider the impact of these thresholds for case definition of hypertension in terms of increased prevalence and treatment thereof.

In this systematic review, for the first time, we attempted to provide an estimate of the mean arterial BP in patients with periodontitis vs. controls. Very interestingly, more than 80% of the included studies reporting levels of blood pressure showed consistently increased levels of systolic and diastolic BP in patients with periodontitis. Further, the exploratory meta-analysis B revealed that patients with periodontitis showed a higher WMD of 4.49 mmHg of SBP and of 2.03 mmHg of DBP. If confirmed in long-term longitudinal studies, periodontitis could represent a novel modifiable risk factor for hypertension at the same strength of diabetes and smoking.[73,74] However, as periodontitis, diabetes and hypertension share common risk factors (such as aging, smoking, and disadvantageous socioeconomic status, among others), residual confounding could affect the magnitude of these associations. It is important to state that this association could also be driven by an association between arterial blood pressure changes and other undetected sources/chronic infections. Further research in identifying the interplay between triggers/bacterial burdens in each individual and their relative contribution on blood pressure is needed.

Raised arterial blood pressure observed in periodontitis could also explain the moderate but consistent higher risk of CV events (i.e. MI and stroke) reported by several investigators in patients with periodontitis when compared to controls.[17] Indeed, an average increase of 5 mmHg of SBP has been consistently associated with a 25% increased mortality from ischaemic heart disease and stroke.[75] These assumptions should all be interpreted with caution because of the high heterogeneity observed in the reported scientific evidence. In particular, varying case definitions of periodontitis and hypertension could have undermined the validity of these observations. Nevertheless, due to the high prevalence of both conditions, the clinical implications for public health systems could be very significant.

This systematic review also confirmed a potential positive effect of treating periodontal inflammation on arterial blood pressure. Inconclusive findings were identified in the selected studies, with only 5 out of 12 intervention trials showing a reduction of SBP/DBP in patients with periodontitis.

Only one RCT was designed to address the question whether non-surgical periodontal therapy could result in reduced arterial BP levels.[65] These authors assessed changes in blood pressure as their primary outcome following non-surgical periodontal therapy. They included 107 pre-hypertensive participants and reported an absolute difference of SBP = 12.57 mmHg 95% CI: 10.45–14.69, $P < 0.05$ and of DBP = 9.65 mmHg 95% CI: 7.06–12.24, $P < 0.05$ after periodontal therapy. As treatment of hypertension has been repeatedly advocated as a key intervention to improve general health, quality of life, and reduce CV complications,[76] periodontitis treatment could represent a novel non-pharmacological therapy to prevent/help manage hypertension. A meta-analysis of RCTs quantified a reduction of 25–30% of coronary heart disease events such as stroke and heart failure with a 10 mmHg reduction in SBP or a 5 mmHg reduction in DBP following anti-hypertensive drug

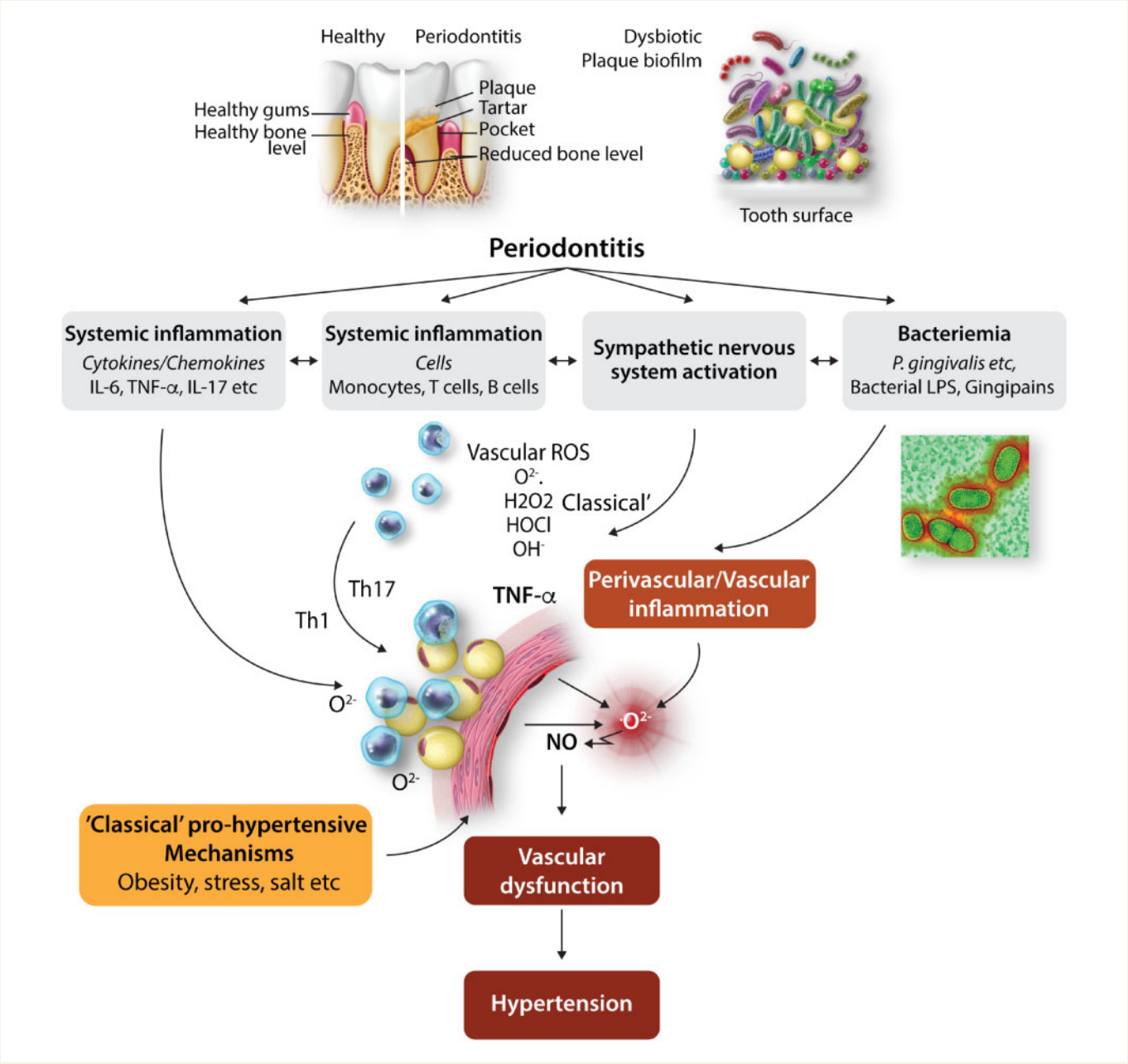


Figure 6 Some pathways through which periodontitis could lead to hypertension and vascular dysfunction. Potential mechanisms causally linking periodontitis with hypertension and vascular dysfunction. NO, nitric oxide; O_2^-, S, superoxide; Th, T-helper cells; ROS, reactive oxygen species.

therapy.[77] Future research should address the hypothesis of the treatment of periodontitis could achieve similar reduction in arterial BP and CV outcomes.

The identification of periodontitis as a possible risk factor for hypertension could be explained by a number of plausible mechanisms (*Figure 6*). Firstly, periodontitis is associated with systemic inflammation, mediators of which, including CRP, IL-6; TNF-α can all affect endothelial function. Clinical evidence suggests periodontitis affects systemic endothelial function and in turn this could impact on hypertension. Our group previously demonstrated that treatment of severe periodontitis improves endothelial function by reduction in systemic inflammation in patients with and without other comorbidities like diabetes.[22,64] Secondly, some reports suggest possible direct effects of oral microbiota related bacteraemia in mediating vascular dysfunction as well. Emerging experimental animal evidence indicates that an immune response to a common periodontal pathogen: *Porphyromonas gingivalis* (*Pg*) results in elevation of BP, vascular inflammation, and endothelial dysfunction.[78] Another possibility may be that cells, including T cells, B cells, and monocyte/macrophages, primed in inflamed periodontium may be more prone to chemotactic recruitment to perivascular adipose tissue and adventitia, a step that has been shown to precede development of vascular dysfunction, hypertension, and atherosclerosis.[79,80] This review therefore raises an important question regarding the causal nature of the association between periodontitis and hypertension.

4.1 Strengths and weaknesses

This systematic review was designed to comprehensively investigate the possible role of periodontitis as a possible novel risk factor for hypertension. A number of limitations however should be highlighted starting with the limited value of systematic reviews of observational studies for ascertaining causality.[81] Moreover, observational studies have intrinsic biases (mostly selection and information bias), hence the results of this systematic review should be interpreted within the context of the methodology used. Nevertheless, this review was broad and inclusive of not only observational but also interventional studies. Besides, because of the link between periodontitis and cardiometabolic risk factors,[17,82] this review also included data from observational studies on MetS and CVD but the authors acknowledge that some of the data may have been missed due to the difficulties in identifying the outcomes within the published reports. Moreover, studies looking at hypertension have inherent problem of the effect of blood pressure measurement technique on the outcome as well as variable degree of reporting of actual criteria of hypertension. Therefore, we have focused on a clear definition of hypertension based mainly on blood pressure values and anti-hypertensive medications. With the exception of a single study[29] most studies have used office rather than ambulatory blood pressure; our quantitative analysis of the effect of periodontitis on blood pressure values adds to the strength of the selected evidence. This study was a pilot intervention trial including only 26 patients with refractory hypertension and periodontitis and the effects of non-surgical periodontal therapy on both systolic and diastolic blood pressure were of greater magnitude of those reported in the other intervention studies. We urge caution in interpreting these results especially in view of the limited sample size and inclusion criteria adopted by the authors.[29] Future intervention trials should all be designed according to appropriate power calculation to determine sample size and include ambulatory blood pressure levels.

One of challenges encountered was to establish the direction of association when studies were included in the quantitative analyses (i.e. dependent and independent variables in the model). This was mainly due to unclear description in the published manuscripts. When a consensus could not be achieved among the reviewers (E.M.A. and J.S.), a third author was consulted (F.D.) or attempts were made to contact the authors for clarification.[43] Another important limitation of this systematic review is the high level of heterogeneity in the case definitions for both, periodontitis and hypertension.[34,83] To overcome this, data were further analysed according to an arbitrary level of confidence in a given case definition of periodontitis. In fact, when an arbitrary confident diagnosis was confirmed, the observed magnitude of association between periodontitis and hypertension was greater. The lack of consistent measures of case definition and severity of periodontitis in the retrieved evidence did not allow for a relevant analysis of extent and severity of periodontitis with all endpoints of blood pressure. We hope in the future reporting of periodontitis is more consistent and allow for such analyses. Lastly, it has been reported that anti-hypertensives such as calcium channel blockers can cause gingival enlargement in 6.3–83% of patients,[84] which should not be confounded with periodontitis.

5. Conclusions

Periodontitis could be associated with increased risk of hypertension in a linear fashion. Further, management of periodontitis could impact on the management of hypertension. Our findings highlight the potential to improve CV outcomes by addressing poor oral health in the general population. Longer and larger studies are needed however to determine whether periodontal treatment benefit patients in terms of CV health, ultimately resulting in reduced morbidity and mortality.

Translational implications

- To raise awareness of the association between periodontitis and hypertension.
- Patients with periodontitis should be informed by oral health professionals of the risk of developing hypertension.
- Oral health advice should be given to all patients with hypertension.
- Prevention and management of periodontitis improves oral/overall health and quality of life and could prevent/improve hypertension.
- Larger observational studies should include internationally recognized case definitions for periodontitis and hypertension.
- Larger and long-term RCTs with reduction of blood pressure as primary outcome should be performed.
- Patient reported outcome measures relevant to hypertension and periodontitis should be included within future study designs.

Supplementary material

Supplementary material is available at *Cardiovascular Research* online.

Acknowledgements

We would like to acknowledge that contribution of this work was undertaken at UCLH/UCL who received a proportion of funding from the Department of Health's NIHR Biomedical Research Centre funding scheme.

Conflict of interest: none declared.

Funding

T.J.G. is funded by European Research Council (ERC) InflammaTENSION project.

References

1. Williams B, Mancia G, Spiering W, Agabiti Rosei E, Azizi M, Burnier M, Clement DL, Coca A, de Simone G, Dominiczak A, Kahan T, Mahfoud F, Redon J, Ruilope L, Zanchetti A, Kerins M, Kjeldsen SE, Kreutz R, Laurent S, Lip GYH, McManus R, Narkiewicz K, Ruschitzka F, Schmieder RE, Shlyakhto E, Tsioufis C, Aboyans V, Desormais I; Authors/Task Force Members. 2018 ESC/ESH Guidelines for the management of arterial hypertension: the Task Force for the management of arterial hypertension of the European Society of Cardiology and the European Society of Hypertension. *J Hypertens* 2018;**36**:1953–2041.
2. Mancia G, Fagard R, Narkiewicz K, Redon J, Zanchetti A, Böhm M, Christiaens T, Cifkova R, De Backer G, Dominiczak A, Galderisi M, Grobbee DE, Jaarsma T, Kirchhof P, Kjeldsen SE, Laurent S, Manolis AJ, Nilsson PM, Ruilope LM, Schmieder RE, Sirnes PA, Sleight P, Viigimaa M, Waeber B, Zannad F. 2013 ESH/ESC Guidelines for the management of arterial hypertension: the Task Force for the management of arterial hypertension of the European Society of Hypertension (ESH) and of the European Society of Cardiology (ESC). *Blood Press* 2013;**22**:193–278.
3. Bath P, Chalmers J, Powers W, Beilin L, Davis S, Lenfant C, Mancia G, Neal B, Whitworth J, Zanchetti A. International Society of Hypertension (ISH): statement on the management of blood pressure in acute stroke. *J Hypertens* 2003;**21**:665.
4. Mancia G, Laurent S, Agabiti-Rosei E, Ambrosioni E, Burnier M, Caulfield MJ, Cifkova R, Clément D, Coca A, Dominiczak A, Erdine S, Fagard R, Farsang C, Grassi G, Haller H, Heagerty A, Kjeldsen SE, Kiowski W, Mallion JM, Manolis A, Narkiewicz K, Nilsson P, Olsen MH, Rahn KH, Redon J, Rodicio J, Ruilope L, Schmieder RE, Struijker-Boudier HAJ, Van Zwieten PA, Viigimaa M, Zanchetti A. Reappraisal of European guidelines on hypertension management: a European Society of Hypertension Task Force document. *Blood Press* 2009;**18**:308–347.

5. Bangalore S, Toklu B, Gianos E, Schwartzbard A, Weintraub H, Ogedegbe G, Messerli FH. Optimal systolic blood pressure target after SPRINT: insights from a network meta-analysis of randomized trials. *Am J Med* 2017;**130**:707–719. e708.
6. Bundy JD, Li C, Stuchlik P, Bu X, Kelly TN, Mills KT, He H, Chen J, Whelton PK, He J. Systolic blood pressure reduction and risk of cardiovascular disease and mortality: a systematic review and network meta-analysis. *JAMA Cardiol* 2017;**2**:775–781.
7. Joffres M, Falaschetti E, Gillespie C, Robitaille C, Loustalot F, Poulter N, McAlister FA, Johansen H, Baclic O, Campbell N. Hypertension prevalence, awareness, treatment and control in national surveys from England, the USA and Canada, and correlation with stroke and ischaemic heart disease mortality: a cross-sectional study. *BMJ Open* 2013;**3**:e003423.
8. Carretero OA, Oparil S. Essential hypertension part I: definition and etiology. *Circulation* 2000;**101**:329–335.
9. Czopek A, Moorhouse R, Guyonnet L, Farrah T, Lenoir O, Owen E, van Bragt J, Costello HM, Menolascina F, Baudrie V, Webb DJ, Kluth DC, Bailey MA, Tharaux P-L, Dhaun N. A novel role for myeloid endothelin-B receptors in hypertension. *Eur Heart J* 2019;**40**:768.
10. Guzik TJ, Hoch NE, Brown KA, McCann LA, Rahman A, Dikalov S, Goronzy J, Weyand C, Harrison DG. Role of the T cell in the genesis of angiotensin II-induced hypertension and vascular dysfunction. *J Exp Med* 2007;**204**:2449–2460.
11. Itani HA, McMaster WG, Saleh MA, Nazarewicz RR, Mikolajczyk TP, Kaszuba AM, Konior A, Prejbisz A, Januszewicz A, Norlander AE, Chen W, Bonami RH, Marshall AF, Poffenberger G, Weyand CM, Madhur MS, Moore DJ, Harrison DG, Guzik TJ. Activation of human T cells in hypertension: studies of humanized mice and hypertensive humans. *Hypertension* 2016;**68**:123–132.
12. Drummond GR, Vinh A, Guzik TJ, Sobey CG. Immune mechanisms of hypertension. *Nat Rev Immunol* 2019;**19**:517–532.
13. Buset S, Walter C, Friedmann A, Weiger R, Borgnakke WS, Zitzmann NU. Are periodontal diseases really silent? A systematic review of their effect on quality of life. *J Clin Periodontol* 2016;**43**:333.
14. Sanz M, D'Aiuto F, Deanfield J, Fernandez AF. European workshop in periodontal health and cardiovascular disease—scientific evidence on the association between periodontal and cardiovascular diseases: a review of the literature. *Eur Heart J Suppl* 2010;**12**:B3–B12.
15. Kassebaum NJ, Bernabé E, Dahiya M, Bhandari B, Murray CJL, Marcenes W. Global burden of severe periodontitis in 1990–2010: a systematic review and meta-regression. *J Dent Res* 2014;**93**:1045–1053.
16. Baehni P, Tonetti MS. Conclusions and consensus statements on periodontal health, policy and education in Europe: a call for action–consensus view 1. *Eur J Dent Educ* 2010;**14**:2–3.
17. Tonetti MS, Dyke TE. Periodontitis and atherosclerotic cardiovascular disease: consensus report of the Joint EFP/AAP Workshop on Periodontitis and Systemic Diseases. *J Clin Periodontol* 2013;**40**:S24.
18. Demmer RT, Papapanou PN. Epidemiologic patterns of chronic and aggressive periodontitis. *Periodontol 2000* 2010;**53**:28–44.
19. Southerland JH. Periodontitis may contribute to poor control of hypertension in older adults. *J Evid Based Dent Pract* 2013;**13**:125–127.
20. Dominy SS, Lynch C, Ermini F, Benedyk M, Marczyk A, Konradi A, Nguyen M, Haditsch U, Raha D, Griffin C, Holsinger LJ, Arastu-Kapur S, Kaba S, Lee A, Ryder MI, Potempa B, Mydel P, Hellvard A, Adamowicz K, Hasturk H, Walker GD, Reynolds EC, Faull RLM, Curtis MA, Dragunow M, Potempa J. *Porphyromonas gingivalis* in Alzheimer's disease brains: evidence for disease causation and treatment with small-molecule inhibitors. *Sci Adv* 2019;**5**:eaau3333.
21. Hujoel PP, Drangsholt M, Spiekerman C, Derouen TA. Examining the link between coronary heart disease and the elimination of chronic dental infections. *J Am Dent Assoc* 2001;**132**:883–889.
22. Tonetti MS, D'aiuto F, Nibali L, Donald A, Storry C, Parkar M, Suvan J, Hingorani AD, Vallance P, Deanfield J. Treatment of periodontitis and endothelial function. *N Engl J Med* 2007;**356**:911–920.
23. Orlandi M, Suvan J, Petrie A, Donos N, Masi S, Hingorani A, Deanfield J, D'Aiuto F. Association between periodontal disease and its treatment, flow-mediated dilatation and carotid intima-media thickness: a systematic review and meta-analysis. *Atherosclerosis* 2014;**236**:39–46.
24. Tsakos G, Sabbah W, Hingorani AD, Netuveli G, Donos N, Watt RG, D'Aiuto F. Is periodontal inflammation associated with raised blood pressure? Evidence from a National US survey. *J Hypertens* 2010;**28**:2386–2393.
25. Tsioufis C, Kasiakogias A, Thomopoulos C, Stefanadis C. Periodontitis and blood pressure: the concept of dental hypertension. *Atherosclerosis* 2011;**219**:1–9.
26. Zeigler CC, Wondimu B, Marcus C, Modéer T. Pathological periodontal pockets are associated with raised diastolic blood pressure in obese adolescents. *BMC Oral Health* 2015;**15**:1.
27. Martin-Cabezas R, Seelam N, Petit C, Agossa K, Gaertner S, Tenenbaum H, Davideau J-L, Huck O. Association between periodontitis and arterial hypertension: a systematic review and meta-analysis. *Am Heart J* 2016;**180**:98–112.
28. D'Aiuto F, Parkar M, Nibali L, Suvan J, Lessem J, Tonetti MS. Periodontal infections cause changes in traditional and novel cardiovascular risk factors: results from a randomized controlled clinical trial. *Am Heart J* 2006;**151**:977–984.
29. Vidal F, Cordovil I, Figueredo CMS, Fischer RG. Non-surgical periodontal treatment reduces cardiovascular risk in refractory hypertensive patients: a pilot study. *J Clin Periodontol* 2013;**40**:681
30. Nibali L, Farias BC, Vajgel A, Tu Y, Donos N. Tooth loss in aggressive periodontitis: a systematic review. *J Dent Res* 2013;**92**:868–875.
31. Armitage GC. Development of a classification system for periodontal diseases and conditions. *Ann Periodontol* 1999;**4**:1–6.
32. Page RC, Eke PI. Case definitions for use in population-based surveillance of periodontitis. *J Periodontol* 2007;**78**:1387–1399.
33. Tonetti M, Claffey N; European Workshop in Periodontology group C. Advances in the progression of periodontitis and proposal of definitions of a periodontitis case and disease progression for use in risk factor research. Group C consensus report of the 5th European workshop in periodontology. *J Clin Periodontol* 2005;**32**: 210–213.
34. Preshaw PM. Definitions of periodontal disease in research. *J Clin Periodontol* 2009;**36**: 1–2.
35. Higgins J, Sterne J, Savović J, Page M, Hróbjartsson A, Boutron I, Reeves B, Eldridge S. A revised tool for assessing risk of bias in randomized trials. *Cochrane Database Syst Rev* 2016;**10**:29–31.
36. Sterne JA, Hernán MA, Reeves BC, Savović J, Berkman ND, Viswanathan M, Henry D, Altman DG, Ansari MT, Boutron I, Carpenter JR, Chan A-W, Churchill R, Deeks JJ, Hróbjartsson A, Kirkham J, Jüni P, Loke YK, Pigott TD, Ramsay CR, Regidor D, Rothstein HR, Sandhu L, Santaguida PL, Schünemann HJ, Shea B, Shrier I, Tugwell P, Turner L, Valentine JC, Waddington H, Waters E, Wells GA, Whiting PF, Higgins JP. ROBINS-I: a tool for assessing risk of bias in non-randomised studies of interventions. *BMJ* 2016;**355**:i4919.
37. Wells G, Shea B, O'connell D, Peterson J, Welch V, Losos M, Tugwell P. *The Newcastle–Ottawa Scale (NOS) for Assessing the Quality of Nonrandomised Studies in Meta-Analyses*. Ottawa (ON): Ottawa Hospital Research Institute; 2009.
38. Begg CB, Mazumdar M. Operating characteristics of a rank correlation test for publication bias. *Biometrics* 1994;**50**:1088–1101.
39. Egger M, Smith GD, Schneider M, Minder C. Bias in meta-analysis detected by a simple, graphical test. *BMJ* 1997;**315**:629–634.
40. Newman MG, Weyant R, Hujoel P. JEBDP improves grading system and adopts strength of recommendation taxonomy grading (SORT) for guidelines and systematic reviews. *J Evid Based Dent Pract* 2007;**7**:147–150.
41. Chobanian AV, Bakris GL, Black HR, Cushman WC, Green LA, Izzo JL Jr, Jones DW, Materson BJ, Oparil S, Wright JT Jr. The seventh report of the joint national committee on prevention, detection, evaluation, and treatment of high blood pressure: the JNC 7 report. *JAMA* 2003;**289**:2560–2571.
42. Grundy SM, Cleeman JI, Daniels SR, Donato KA, Eckel RH, Franklin BA, Gordon DJ, Krauss RM, Savage PJ, Smith SC, Spertus JA, Costa F. Diagnosis and management of the metabolic syndrome: an American Heart Association/National Heart, Lung, and Blood Institute scientific statement. *Circulation* 2005;**112**:2735–2752.
43. Boland MR, Hripcsak G, Albers DJ, Wei Y, Wilcox AB, Wei J, Li J, Lin S, Breene M, Myers R, Zimmerman J, Papapanou PN, Weng C. Discovering medical conditions associated with periodontitis using linked electronic health records. *J Clin Periodontol* 2013;**40**:474–482.
44. Taylor B, Tofler G, Morel-Kopp M-C, Carey H, Carter T, Elliott M, Dailey C, Villata L, Ward C, Woodward M, Schenck K. The effect of initial treatment of periodontitis on systemic markers of inflammation and cardiovascular risk: a randomized controlled trial. *Eur J Oral Sci* 2010;**118**:350
45. Lee J-H, Lee J-S, Park J-Y, Choi J-K, Kim D-W, Kim Y-T, Choi S-H. Association of lifestyle-related comorbidities with periodontitis: a nationwide cohort study in Korea. *Medicine* 2015;**94**:e1567. Erratum in: Medicine 2016;95:e365d.
46. D'Aiuto F, Sabbah W, Netuveli G, Donos N, Hingorani AD, Deanfield J, Tsakos G. Association of the metabolic syndrome with severe periodontitis in a large US population-based survey. *J Clin Endocrinol Metab* 2008;**93**:3989–3994.
47. Tu YK, D'aiuto F, Lin HJ, Chen YW, Chien KL. Relationship between metabolic syndrome and diagnoses of periodontal diseases among participants in a large Taiwanese cohort. *J Clin Periodontol* 2013;**40**:994–1000.
48. Rivas-Tumanyan S, Spiegelman D, Curhan GC, Forman JP, Joshipura KJ. Periodontal disease and incidence of hypertension in the health professionals follow-up study. *Am J Hypertens* 2012;**25**:770–776.
49. Morita T, Yamazaki Y, Mita A, Takada K, Seto M, Nishinoue N, Sasaki Y, Motohashi M, Maeno M. A cohort study on the association between periodontal disease and the development of metabolic syndrome. *J Periodontol* 2010;**81**:512–519.
50. Tuominen R, Reunanen A, Paunio M, Paunio I, Aromaa A. Oral health indicators poorly predict coronary heart disease deaths. *J Dent Res* 2003;**82**:713–718.
51. Desvarieux M, Demmer RT, Jacobs DR Jr, Rundek T, Boden-Albala B, Sacco RL, Papapanou PN. Periodontal bacteria and hypertension: the oral infections and vascular disease epidemiology study (INVEST). *J Hypertens* 2010;**28**:1413.
52. Gomes-Filho IS, das Mercês MC, de Santana Passos-Soares J, Seixas da Cruz S, Teixeira Ladeia AM, Trindade SC, de Moraes Marcílio Cerqueira E, Freitas Coelho JM, Marques Monteiro FM, Barreto ML, Pereira Vianna MI, Nascimento Costa MDC, Seymour GJ, Scannapieco FA. Severity of periodontitis and metabolic syndrome: is there an association? *J Periodontol* 2016;**87**:357–366.

53. Han DH, Lim SY, Sun BC, Paek D, Kim HD. The association of metabolic syndrome with periodontal disease is confounded by age and smoking in a Korean population: the Shiwha–Banwol environmental health study. *J Clin Periodontol* 2010; **37**:609–616.
54. Gordon JH, LaMonte MJ, Genco RJ, Zhao J, Cimato TR, Hovey KM, Wactawski-Wende J. Association of clinical measures of periodontal disease with blood pressure and hypertension among postmenopausal women. *J Periodontol* 2018;**89**:1193–1202.
55. Türkoğlu O, Barış N, Tervahartiala T, Şenarslan Ö, Sorsa T, Atilla G. Evaluation of systemic levels of neutrophilic enzymes in patients with hypertension and chronic periodontitis. *J Periodontol* 2014;**85**:908–916.
56. Khocht A, Rogers T, Janal M, Brown M. Gingival fluid inflammatory biomarkers and hypertension in African Americans. *JDR Clin Trans Res* 2017;**2**:269–277.
57. Albush MM, Razan KK, Al Dieri MR. Effect of surgical and non-surgical periodontal debridement on vascular thrombotic markers in hypertensives. *J Indian Soc Periodontol* 2013;**17**:324.
58. Graziani F, Cei S, Tonetti M, Paolantonio M, Serio R, Sammartino G, Gabriele M, D'aiuto F. Systemic inflammation following non-surgical and surgical periodontal therapy. *J Clin Periodontol* 2010;**37**:848
59. Higashi Y, Goto C, Jitsuiki D, Umemura T, Nishioka K, Hidaka T, Takemoto H, Nakamura S, Soga J, Chayama K, Yoshizumi M, Taguchi A. Periodontal infection is associated with endothelial dysfunction in healthy subjects and hypertensive patients. *Hypertension* 2008;**51**:446–453.
60. Higashi Y, Goto C, Hidaka T, Soga J, Nakamura S, Fujii Y, Hata T, Idei N, Fujimura N, Chayama K, Kihara Y, Taguchi A. Oral infection-inflammatory pathway, periodontitis, is a risk factor for endothelial dysfunction in patients with coronary artery disease. *Atherosclerosis* 2009;**206**:604–610.
61. Houcken W, Teeuw WJ, Bizzarro S, Rodriguez EA, Mulders TA, van den Born BJH, Loos BG. Arterial stiffness in periodontitis patients and controls. A case–control and pilot intervention study. *J Hum Hypertens* 2016;**30**:24–29.
62. Seinost G, Wimmer G, Skerget M, Thaller E, Brodmann M, Gasser R, Bratschko RO, Pilger E. Periodontal treatment improves endothelial dysfunction in patients with severe periodontitis. *Am Heart J* 2005;**149**:1050–1054.
63. López NJ, Quintero A, Casanova PA, Ibieta CI, Baelum V, López R. Effects of periodontal therapy on systemic markers of inflammation in patients with metabolic syndrome: a controlled clinical trial. *J Periodontol* 2012;**83**:267–278.
64. D'Aiuto F, Gkranias N, Bhowruth D, Khan T, Orlandi M, Suvan J, Masi S, Tsakos G, Hurel S, Hingorani AD. Systemic effects of periodontitis treatment in patients with type 2 diabetes: a 12 month, single-centre, investigator-masked, randomised trial. *Lancet Diabetes Endocrinol* 2018;**6**:954–965.
65. Zhou Q-B, Xia W-H, Ren J, Yu B-B, Tong X-Z, Chen Y-B, Chen S, Feng L, Dai J, Tao J, Yang J-Y. Effect of intensive periodontal therapy on blood pressure and endothelial microparticles in patients with prehypertension and periodontitis: a randomized controlled trial. *J Periodontol* 2017;**88**:711–722.
66. Kawabata Y, Ekuni D, Miyai H, Kataoka K, Yamane M, Mizutani S, Irie K, Azuma T, Tomofuji T, Iwasaki Y, Morita M. Relationship between prehypertension/hypertension and periodontal disease: a prospective cohort study. *Am J Hypertens* 2016;**29**: 388–396.
67. Morita T, Yamazaki Y, Fujiharu C, Ishii T, Seto M, Nishinoue N, Sasaki Y, Nakai K, Tanaka H, Kawato T, Maeno M. Association between the duration of periodontitis and increased cardiometabolic risk factors: a 9-year cohort study. *Metab Syndr Relat Disord* 2016;**14**:475–482.
68. Lee J-H, Oh J-Y, Youk T-M, Jeong S-N, Kim Y-T, Choi S-H. Association between periodontal disease and non-communicable diseases: a 12-year longitudinal health-examinee cohort study in South Korea. *Medicine* 2017;**96**:e7398.
69. Bonato C, do-Amaral C, Belini L, Salzedas LMP, Oliveira S. Hypertension favors the inflammatory process in rats with experimentally induced periodontitis. *J Periodontal Res* 2012;**47**:783
70. Demmer RT, Papapanou PN, Jacobs DR Jr, Desvarieux M. Bleeding on probing differentially relates to bacterial profiles: the oral infections and vascular disease epidemiology study. *J Clin Periodontol* 2008;**35**:479–486.
71. Whelton PK, Carey RM, Aronow WS, Casey DE, Collins KJ, Himmelfarb CD, DePalma SM, Gidding S, Jamerson KA, Jones DW. 2017 ACC/AHA/AAPA/ABC/ACPM/AGS/APhA/ASH/ASPC/NMA/PCNA guideline for the prevention, detection, evaluation, and management of high blood pressure in adults: a report of the American College of Cardiology/American Heart Association Task Force on Clinical Practice Guidelines. *J Am Coll Cardiol* 2018;**71**:e127–e248.
72. Khera R, Lu Y, Lu J, Saxena A, Nasir K, Jiang L, Krumholz HM. Impact of 2017 ACC/AHA guidelines on prevalence of hypertension and eligibility for antihypertensive treatment in United States and China: nationally representative cross sectional study. *BMJ* 2018;**362**:k2357.
73. Cheung BM, Li C. Diabetes and hypertension: is there a common metabolic pathway? *Curr Atheroscler Rep* 2012;**14**:160–166.
74. Saladini F, Benetti E, Fania C, Mos L, Casiglia E, Palatini P. Effects of smoking on central blood pressure and pressure amplification in hypertension of the young. *Vasc Med* 2016;**21**:422–428.
75. Lewington S, Clarke R, Qizilbash N, Peto R, Collins R; Prospective Studies Collaboration. Age-specific relevance of usual blood pressure to vascular mortality: a meta-analysis of individual data for one million adults in 61 prospective studies. *Lancet* 2002;**360**:1903–1913.
76. Ettehad D, Emdin CA, Kiran A, Anderson SG, Callender T, Emberson J, Chalmers J, Rodgers A, Rahimi K. Blood pressure lowering for prevention of cardiovascular disease and death: a systematic review and meta-analysis. *Lancet* 2016;**387**:957–967.
77. Law M, Morris J, Wald N. Use of blood pressure lowering drugs in the prevention of cardiovascular disease: meta-analysis of 147 randomised trials in the context of expectations from prospective epidemiological studies. *BMJ* 2009;**338**:b1665.
78. Czesnikiewicz-Guzik M, Nosalski R, Mikolajczyk TP, Vidler F, Dohnal T, Dembowska E, Graham D, Harrison DG, Guzik TJ. Th1-type immune responses to *Porphyromonas gingivalis* antigens exacerbate angiotensin II-dependent hypertension and vascular dysfunction. *Br J Pharmacol* 2019;**176**:1922–1931.
79. Guzik TJ, Skiba DS, Touyz RM, Harrison DG. The role of infiltrating immune cells in dysfunctional adipose tissue. *Cardiovasc Res* 2017;**113**:1009–1023.
80. Mikolajczyk TP, Nosalski R, Szczepaniak P, Budzyn K, Osmenda G, Skiba D, Sagan A, Wu J, Vinh A, Marvar PJ, Guzik B, Podolec J, Drummond G, Lob HE, Harrison DG, Guzik TJ. Role of chemokine RANTES in the regulation of perivascular inflammation, T-cell accumulation, and vascular dysfunction in hypertension. *FASEB J* 2016;**30**: 1987–1999.
81. Grimes DA, Schulz KF. An overview of clinical research: the lay of the land. *Lancet* 2002;**359**:57–61.
82. Nibali L, Tatarakis N, Needleman I, Tu Y-K, D'aiuto F, Rizzo M, Donos N. Association between metabolic syndrome and periodontitis: a systematic review and meta-analysis. *J Clin Endocrinol Metab* 2013;**98**:913–920.
83. Savage A, Eaton KA, Moles DR, Needleman I. A systematic review of definitions of periodontitis and methods that have been used to identify this disease. *J Clin Periodontol* 2009;**36**:458–467.
84. Hallmon WW, Rossmann JA The role of drugs in the pathogenesis of gingival overgrowth. A collective review of current concepts. *Periodontol 2000* 1999;**21**:176–196.

Appendix 1 Prisma Statement

Table 1| Checklist of items to include when reporting a systematic review or meta-analysis

Section/topic	Item No	Checklist item	Reported on page No
Title			1
Title	1	Identify the report as a systematic review, meta-analysis, or both	
Abstract			
Structured summary	2	Provide a structured summary including, as applicable, background, objectives, data sources, study eligibility criteria, participants, interventions, study appraisal and synthesis methods, results, limitations, conclusions and implications of key findings, systematic review registration number	3
Introduction			
Rationale	3	Describe the rationale for the review in the context of what is already known	5
Objectives	4	Provide an explicit statement of questions being addressed with reference to participants, interventions, comparisons, outcomes, and study design (PICOS)	6
Methods			
Protocol and registration	5	Indicate if a review protocol exists, if and where it can be accessed (such as web address), and, if available, provide registration information including registration number	6
Eligibility criteria	6	Specify study characteristics (such as PICOS, length of follow-up) and report characteristics (such as years considered, language, publication status) used as criteria for eligibility, giving rationale	7
Information sources	7	Describe all information sources (such as databases with dates of coverage, contact with study authors to identify additional studies) in the search and date last searched	8
Search	8	Present full electronic search strategy for at least one database, including any limits used, such that it could be repeated	7
Study selection	9	State the process for selecting studies (that is, screening, eligibility, included in systematic review, and, if applicable, included in the meta-analysis)	8
Data collection process	10	Describe method of data extraction from reports (such as piloted forms, independently, in duplicate) and any processes for obtaining and confirming data from investigators	8
Data items	11	List and define all variables for which data were sought (such as PICOS, funding sources) and any assumptions and simplifications made	8,9
Risk of bias in individual studies	12	Describe methods used for assessing risk of bias of individual studies (including specification of whether this was done at the study or outcome level), and how this information is to be used in any data synthesis	9
Summary measures	13	State the principal summary measures (such as risk ratio, difference in means).	9
Synthesis of results	14	Describe the methods of handling data and combining results of studies, if done, including measures of consistency (such as I^2 statistic) for each meta-analysis	9, 10
Risk of bias across studies	15	Specify any assessment of risk of bias that may affect the cumulative evidence (such as publication bias, selective reporting within studies)	9
Additional analyses	16	Describe methods of additional analyses (such as sensitivity or subgroup analyses, meta-regression), if done, indicating which were pre-specified	10
Results			
Study selection	17	Give numbers of studies screened, assessed for eligibility, and included in the review, with reasons for exclusions at each stage, ideally with a flow diagram	10
Study characteristics	18	For each study, present characteristics for which data were extracted (such as study size, PICOS, follow-up period) and provide the citations	Appendix 3
Risk of bias within studies	19	Present data on risk of bias of each study and, if available, any outcome-level assessment (see item 12).	Appendix 4
Results of individual studies	20	For all outcomes considered (benefits or harms), present for each study (a) simple summary data for each intervention group and (b) effect estimates and confidence intervals, ideally with a forest plot	Figures 2-8
Synthesis of results	21	Present results of each meta-analysis done, including confidence intervals and measures of consistency	Figures 2-8
Risk of bias across studies	22	Present results of any assessment of risk of bias across studies (see item 15)	Page 14, appendix 4
Additional analysis	23	Give results of additional analyses, if done (such as sensitivity or subgroup analyses, meta-regression) (see item 16)	11, 12
Discussion			
Summary of evidence	24	Summarise the main findings including the strength of evidence for each main outcome; consider their relevance to key groups (such as health care providers, users, and policy makers)	14-16
Limitations	25	Discuss limitations at study and outcome level (such as risk of bias), and at review level (such as incomplete retrieval of identified research, reporting bias)	16-17

Table 1 (continued)

Section/topic	Item No	Checklist item	Reported on page No
Conclusions	26	Provide a general interpretation of the results in the context of other evidence, and implications for future research	17
Funding			
Funding	27	Describe sources of funding for the systematic review and other support (such as supply of data) and role of funders for the systematic review	2

Appendix 2. Search Strategy

Medline	Embase	Central	Web of Science	Lilacs
#1 exp periodontal diseases/ #2 periodont$.mp. #3 gingiv$.mp. #4 ((blood or bleed$) adj4 prob$).mp. #5 (ging$ adj disease).mp. #6 exp periodontics/ #7 or/1-6 #8 exp hypertension/ #9 exp blood pressure/ #10 hypertens$.mp. #11 ((elevat$ or high$ or rais$) adj3 (diastolic or systolic or arterial or blood) adj pressure).mp. #12 bloodpressure.mp. #13 or/8-12 #14 7 and 13 #15 exp animals/ not humans.sh. #16 14 not 15	#1 periodont$.mp. #2 gingiv$.mp. #3 ((blood or bleed$) adj4 prob$).mp. #4 (ging$ adj disease).mp. #5 exp periodontal disease/ #6 exp periodontics/ #7 1 or 2 or 3 or 4 or 5 or 6 #8 hypertens$.mp. #9 ((elevat$ or high$ or rais$) adj3 (diastolic or systolic or arterial or blood) adj pressure).mp. #10 bloodpressure.mp. #11 exp hypertension/ #12 exp blood pressure/ #13 8 or 9 or 10 or 11 or 12 #14 7 and 13 #15 animal/ #16 human/ #17 15 not (15 and 16) #18 14 not 17 #19 Clinical trial/ #20 Randomized controlled trial/ #21 Randomization/ #22 Single blind procedure/ #23 Double blind procedure/ #24 Crossover procedure/ #25 Placebo/ #26 Randomi?ed controlled trial$.tw. #27 Rct.tw. #28 Random allocation.tw.	#1 MeSH descriptor: [Periodontal Diseases] explode all trees #2 MeSH descriptor: [Periodontics] explode all trees #3 periodont* #4 gingiv* #5 (blood or bleed*) next/4 prob* #6 ging* next disease #7 #1 or #2 or #3 or #4 or #5 or #6 #8 MeSH descriptor: [Hypertension] explode all trees #9 MeSH descriptor: [Blood Pressure] explode all trees #10 hypertens* #11 (elevat* or high* or rais*) next/3 (diastolic or systolic or arterial or blood) next pressure #12 bloodpressure #13 #8 or #9 or #10 or #11 or #12 #14 #7 and #13	# 1 (periodont*) # 2 (gingiv*) # 3 ((blood or bleed*) next/4 prob*) # 4 (ging* next disease) # 5 #4 OR #3 OR #2 OR #1 # 6 (hypertens*) # 7 (((elevat* or high* or rais*) next/3 (diastolic or systolic or arterial or blood) next pressure)) # 8 (bloodpressure) # 9 #8 OR #7 OR #6 #10 #9 AND #5	Subject descriptor: (periodontal diseases or periodontics) and (hypertension or blood pressure) Or Word: ((periodont$ or gingiv$ or ((blood or bleed$) and prob$) or (ging$ and disease)) and (hypertens$ or ((elevat$ or high$ or rais$) and (diastolic or systolic or arterial or blood) and pressure)) or bloodpressure)

	#29 Randomly allocated.tw. #30 Allocated randomly.tw. #31 (allocated adj2 random).tw. #32 Single blind$.tw. #33 Double blind$.tw. #34 ((treble or triple) adj blind$).tw. #35 Placebo$.tw. #36 Prospective study/ #37 19 or 20 or 21 or 22 or 23 or 24 or 25 or 26 or 27 or 28 or 29 or 30 or 31 or 32 or 33 or 34 or 35 or 36 #38 Case study/ #39 Case report.tw. #40 Abstract report/ or letter/ #41 38 or 39 or 40 #42 37 not 41 #43 Clinical study/ #44 Case control study/ #45 Family study/ #46 Longitudinal study/ #47 Retrospective study/ #48 Prospective study/ #49 Randomized controlled trials/ #50 48 not 49 #51 Cohort analysis/ #52 (Cohort adj (study or studies)).mp. #53 (Case control adj (study or studies)).tw. #54 (follow up adj (study or studies)).tw. #55 (observational adj (study or studies)).tw.			

	#56 (epidemiologic$ adj (study or studies)).tw. #57 (cross sectional adj (study or studies)).tw. #58 or/43-47,50-57 #59 42 or 58 #60 18 and 59			

Appendix 3. Evidence Tables

EVIDENCE TABLE FOR CROSS-SECTIONAL STUDIES

Author, year, country, title	Population of study	Periodontal criteria	Blood pressure (BP)	Observed effect (expressed as OR, RR, etc…)	Publication conclusions
Ahn *et al.,* (2015) Korea The association of hypertension with periodontitis is highlighted in female adults: results from the fourth Korea national health and nutrition examination survey	N=14,625 participants Age: >19 years. Gender: Female: 8,378, Male: 6,247 Smoking status: reported as never or ever in lifetime Systemic health: Questionnaire + clinical examination by trained staff members	CPITN score of 3 or 4	SBP ≥ 140 mm Hg or DBP ≥ 90 mm Hg or taking antihypertensive medication	Periodontal disease was dependent variable PR= Prevalence Ratio= 1.1 (1.04–1.16) Trend p= 0.001 38.4% of people with HT (≥90/140) (1984 people) had periodontitis (CPITN 3/4) vs 35.2% (1820 people) without HT had periodontitis	HT was associated with periodontitis in Korean female adults independent of known confounders
Alade et al. (2018) Nigeria Association of Elevated C-Reactive Protein with Severe Periodontitis in Hypertensive Patients in Lagos, Nigeria: A Pilot Study	N: 50 hypertensive patients with chronic periodontitis Age: Mean age was 52.4 ± 6.96 years (range 37–73 years) Gender: Female: 39, Male: 11 Smoking status: Excluded smokers Systemic health: Not recorded other than hypertension Serum concentration of CRP was measured in milligrams per litre using the immunoturbidimetry method	Periodontitis: At least one interproximal site with probing depth ≥4 mm Classification of moderate and severe periodontitis: CDC/AAP criteria (Page & Eke, 2007) Mild periodontitis: classified periodontitis that did not fit the first two categories into mild periodontitis	Persistent blood pressure of SBP/DBP ≥140/90 mmHg. (Consecutive hypertensive patients who visited to the cardiology outpatient clinic)	The median CRP levels of participants with mild, moderate, and severe chronic periodontitis were 1.0 (0.6, 2.2), 2.4 (1.1, 4.8), and 4.1 (3.3, 9.4), respectively. The difference between the three groups was statistically significant (P = 0.006)	There was a significant association between the severity of chronic periodontitis in the hypertensive individuals with elevated CRP levels. Thus, increased CRP levels in this category of hypertensive participants may place them at a higher risk for CVD, and this further highlights the need for better collaboration between physicians and dentists.

Author, year, country, title	Population of study	Periodontal criteria	Blood pressure (BP)	Observed effect (expressed as OR, RR, etc…)	Publication conclusions
Al-Emadi *et al.,* (2006) United States Systemic diseases among patients with and without alveolar bone loss	N=460 adults ≥18 years. Mean age: 47 years, Gender: 52% women Smoking status: self-reported (yes/no) Systemic health: self-reported	Radiographic assessment of bone loss: ≥2.5 mm classified as moderate/severe periodontitis, <2.5 mm as no/mild periodontitis	Self-reported systemic diseases from dental records	OR= 4.2, 95% CI (2.2-8.1) risk of HT for patients with moderate to severe chronic periodontitis vs patients with no or mild periodontitis. Adjusted for age, gender, marital status, smoking history, and number of teeth present About 34% of subjects (n=72) with moderate to severe alveolar bone loss reported a history of hypertension, whereas fewer than 8% of patients (n=16) with no or mild alveolar bone loss reported having the disease	Subjects with moderate to severe alveolar bone loss have an increased prevalence of systemic diseases, especially hypertension and diabetes mellitus
Angeli *et al.,* (2003) Italy Association Between Periodontal Disease and Left Ventricle Mass in Essential Hypertension	N= 104 consecutive hypertensive subjects. None of the subjects had been previously treated or screened for hypertension. Mean age: 57± 10 years old Gender: 53% men Smoking status: reported but criteria to define smoking is not mentioned. Systemic health: Patients with untreated HT	CPITN	HT SBP> 140 mm Hg and/or DBP> 90 mm Hg in 3 different visits over 1 month	-Logistic regression analysis: Body surface area, SBP/DBP, interventricular septum, LV posterior wall, LV internal diameter, relative wall thickness, and LV mass showed an association (all, $p<0.05$) with moderate-to-severe periodontal disease. -SBP relationship with moderate-severe periodontitis: $\beta=0.060 \pm 0.015$, $p \leq 0.0001$ -DBP relationship with moderate-severe periodontitis: $\beta=0.054 \pm 0.02$, $p \leq 0.0001$	Findings suggest a direct association between severity of periodontitis and left ventricular mass in subjects with essential HT. Periodontal evaluation might contribute to refine cardiovascular risk assessment in HT

Author, year, country, title	Population of study	Periodontal criteria	Blood pressure (BP)	Observed effect (expressed as OR, RR, etc…)	Publication conclusions
Aoyama et al., (2017) Japan Specific periodontopathic bacterial infection affects hypertension in male patients with cardiovascular disease.	N=611 patients with CVD Age: 61-80 years old. Mean age=70.7y in HT group vs 68.7 in non-HT group Gender: 34% of women in non-HT group and 24% in the HT group. Smoking status: obtained by interview Systemic health: Recorded	Full mouth chart recorded. No diagnosis of periodontal disease established	Physical examination but not specified case definition	PPD of 71–80-year-old men with HT was deeper than that of non-HT subjects (as Fig. 1 in the manuscript). There was no statistical difference of CAL (Fig. 2) -OR periodontal bacteria AA in subgingival plaque→ HT AA: crude OR= 3.20, 95% CI (1.17–8.74), p=0.0125 Adjusted OR= 2.48 95% CI (0.89–8.07), p= 0.0838 For diabetes mellitus, dyslipidaemia, obesity, and low-density lipoprotein cholesterol -OR periodontal bacteria PI in subgingival plaque→ HT Crude: OR=4.46 (1.30–15.3) p=0.005 Adjusted: OR= 4.21 (1.38–18.4) P=0.0095 For diabetes mellitus, dyslipidaemia, and obesity	Specific periodontopathic bacterial infection may affect HT in male CVD patients. Further investigations needed to reveal the detailed causal relationship
Arowojolu *et al.,* (2016) Nigeria An evaluation of the possible relationship between chronic periodontitis and hypertension	N=100 subjects Mean age: 51 ± 13.6 years [20 – 88 years]. Gender: 45% male, male/female ratio of 1:1.2 Smoking status: smokers were excluded from the study. Systemic disease recorded	CPITN Simplified Oral Hygiene Index [OHI-S]	Measured clinically and classified according to the recommendation by Chobanian *et al.,* (2003)	SBP Pearson correlation coefficient to worse CPITN: 0.143 (not statistically significant) DBP Pearson correlation coefficient to worse CPITN: 0.037 and not statistically significant	In view of the significant relationship between the BP and the mean carotid artery intima media thickness, when compared with the OH status of respondents in this study, more emphasis should be laid on regular preventive dental visits as a way of decreasing cardiovascular health risk

Author, year, country, title	Population of study	Periodontal criteria	Blood pressure (BP)	Observed effect (expressed as OR, RR, etc…)	Publication conclusions
Benguigui *et al*., (2010) France Metabolic syndrome, insulin resistance, and periodontitis: a cross-sectional study in a middle-aged French population	N=255 participants of general population participating on national survey (MONA LISA) to estimate the prevalence of cardio-vascular risk factors -No periodontitis (N=54), age 53.0 ± 9.8, 21% males -Moderate periodontitis (N=102), age 59.6 ± 9.8, 54.9% males -Severe periodontitis (N=99), age 58.9±8.2, 63.6% males Smoking status: smokers were divided into three categories: non-smokers, former, and current smokers. All smoking practices were considered (cigarettes, cigarillos, cigars, and pipe) Systemic health: medical history recorded as per face-to face interview	Page & Eke (2007)	BP recorded with sphygmomanometer Omrom 705 HT was defined as SBP ≥140 mm Hg or DBP≥90 mm Hg or use of antihypertensive drugs	The prevalence of HT among participants defined as no periodontitis, moderate or severe periodontitis was 14 (25.9%), 38 (37.2%) and 48 (48.5%) respectively p= 0.021 Mean BP for non-periodontal patients: SBP 124.8mmHg ±16.7 DBP 78.4mmHg ± 9.4 Mean BP for periodontal patients: SBP 135.6 mm Hg ± 19.7 DBP 83.0 mm Hg ±10.3 Adjusted for confounders hypertension with moderate periodontitis OR= 0.83, 95%CI (0.34–1.99) (Not statistically significant) HT with severe periodontitis OR= 1.22; 95%CI (0.50–2.98) (not statistically significant)	The data support the relationships between metabolic disturbances and periodontitis, with a central role of insulin resistance
Beukers *et al.,* (2017) The Netherlands Periodontitis is an independent risk indicator for atherosclerotic cardiovascular diseases among 60174 participants in a large dental school in the Netherlands	N= 60,174 individuals were identified from dental participants at ACTA in this time period (1998–2013). Age: > 36 years Gender: 49.2% of periodontitis patients and 45.4 of non-periodontitis patients Smoking status: Recorded as yes or no Systemic disease: medical health questionnaire; this is verified by interview and entered into the electronic health record by the dental professional	If at least one of the diagnostic and treatment codes for periodontitis (also corresponding to dental care insurance codes) was found in the electronic health record database	Medical health questionnaire	-12.3% of patients with periodontitis had HT versus 4.2% of non-periodontitis patients. The difference and CI= 8.1% (7.4% to 8.7%) -After adjustment for the confounders (sex, SES, age at intake, smoking, diabetes mellitus, hypertension, hypercholesterolaemia), periodontitis remained independently associated with ACVD OR= 1.59; 95% CI (1.39 - 1.81). With subsequent stratification for age and sex, periodontitis remained independently associated with ACVD	This cross-sectional analysis of a large cohort in the Netherlands of 60,174 participants shows the independent association of periodontitis with ACVD

Author, year, country, title	Population of study	Periodontal criteria	Blood pressure (BP)	Observed effect (expressed as OR, RR, etc…)	Publication conclusions
Buhlin *et al.,* (2002) Sweden Oral health and cardiovascular disease in Sweden. Results of a national questionnaire survey	N= 2,839 Age: age groups (years) 20–39= 1,215 (42.8%) 40–59= 833 (29.4%) 60–84= 791 (27.8%) Gender: 1,645 women (57.9%). Smoking status: Non-smoker, former, current smoker. Systemic health: 52 questions about the dental care insurance system, dental care habits, oral health, CVD and the socio-economic variables of income, education, profession and civil status	Questionnaire based on the following questions: -Bleeding gums -Loose teeth -Denture - ‘‘Has somebody told you that you have deep pockets around your teeth?’’	Concerning CVD, the participants were asked if they had had any type of CVD during the last 9 years. If they answered ‘‘yes’’, they were asked to specify the type	Logistic regression: -Bleeding gums/hypertension: OR= 1.77 (1.26–2.48), p=0.001 -Loose teeth/Hypertension: OR= 1.01 (0.62–1.65), p=0.96 -Deep pockets: OR=1.20 (0.82–1.76), p=0.35 -Dentures/Hypertension: OR=1.24 (0.84–1.84), p=0.28 Adjusted for age, gender, smoking, income level, civil status and education	This study shows an association between cardiovascular disease and bleeding gums, and presence of removable dentures. However, it does not indicate the nature of this association. More studies are needed to determine whether the relation between the two diseases is causal or only a co-variation
Goulart *et al.,* (2017) Brazil Relationship between periodontal disease and cardiovascular risk factors among young and middle-aged Brazilians. cross-sectional study	N= 539 subjects without prior cardiovascular disease Gender: 82% male Mean age: 45 years ± 8.8 Smoking status: checked by cardiologists as (never, former and current) Systemic disease: evaluation focusing on cardiologic assessment and clinical examinations	Full mouth chart evaluation Diagnosis of periodontitis: defined as presence of four or more teeth with one or more sites with PPD ≥ 4 mm and CAL ≥ 3mm	Use of medication to treat hypertension, and systolic BP ≥ 140 mmHg, or diastolic BP ≥ 90 mmHg	42.6% of periodontitis patients (29) had hypertension vs 34.1% (93) of gingivitis patients and 28.8% (57) of patients with no periodontal disease. OR hypertension→Perio OR=1.35, 95% CI [0.69-2.66] p=0.39. Adjusted for age, sex, smoking, current alcohol consumption and oral hygiene Unadjusted OR=1.84, 95%CI [1.04-3.26], p=0.04. A positive association with HT was found, but after multivariate adjustments this association was no longer significant. Some characteristics, including the distribution of CVRFs and age, may be associated with these disparities among studies	This study not find any significant associations between cardiovascular risk factors and periodontal disease in this sample

Author, year, country, title	Population of study	Periodontal criteria	Blood pressure (BP)	Observed effect (expressed as OR, RR, etc…)	Publication conclusions
Chauhan *et al.,* (2016) India Correlation of serum and salivary cytokines level with clinical parameters in metabolic syndrome with periodontitis	N=1,222 Age: Percentages given in age groups: A: Chronic PD MS (*N*=25) B: MS (N= 38); C: Chronic PD (N = 34), D: Control (N = 25) Gender: 54.91% male versus 45.09% female Smoking status: history of habit of smoking or chewing Systemic disease: recorded	≥2 or more teeth with PPD ≥4 mm and CAL ≥ 4 mm that bleed on probing (as per Armitage, 1999)	MetS according to the International Diabetes Federation (IDF) criteria for MS	SBP/DBP (Mean±SD) for: -PD group: 126 ± 8.41/ 84 ± 8.7) -Control group: 125.36 ± 3.90/ 81.36 ± 2.87)	Pro-inflammatory marker TNF-α has correlation with clinical parameters in patients of MS having PD. The study suggests level of salivary TNF-α may be utilized as a surrogate marker of MS and PD
Chen *et al.,* (2016) China Metabolic syndrome and periodontal disease among civilian pilots	N=303 civilian pilots Mean age: 34.92 ± 7.66 y Gender: Not mentioned. male? Smoking status: tobacco consumption recorded Systemic disease: Questionnaire and physical examination	CPI ≥ 3 (WHO)	High BP (SBP ≥130 mm Hg or DBP ≥ 85 mmHg)	Significant correlation (0.42) found between BP and CPI ($p < 0.05$).	The prevalence of MetS was sufficiently high to be a matter of medical concern, and was associated with PD among civilian pilots
Choi et *al.,* (2015) Korea Associations among oral hygiene behaviour and hypertension prevalence and control: the 2008 to 2010 Korea National Health and Nutrition Examination Survey	N= 19,560 adults Age: 9,801 men [19 - 91], 9,759 women [19-95] Smoking status: 1) Non-smokers (never smoked or had smoked <100 cigarettes in their lifetime) 2) Ex-smokers. 3) Current smokers. Systemic health: recorded	The (WHO) community periodontal index (CPI)	Hypertension was defined as an average BP ≥140/90 mm Hg or the use of antihypertensive medication	The adjusted OR of HT prevalence was OR= 1.195, CI 95% (1.033 - 1.383) for individuals who brushed their teeth OR for HT as reported in Martin-Cabezas, (2016). OR= 2.34, CI 95% (2.20-2.50)	Individuals with poor OH behaviour are more likely to have a higher prevalence of HT, even before periodontitis is shown. OH behaviour may be considered an independent risk indicator for HT, and maintaining good oral hygiene may help to prevent and control HT

Author, year, country, title	Population of study	Periodontal criteria	Blood pressure (BP)	Observed effect (expressed as OR, RR, etc…)	Publication conclusions
Chrysanthakopoulos & Chrysanthako-poulos (2016) Greece Association between indices of clinically-defined periodontitis and self-reported history of systemic medical conditions	N= 3,360 outpatients Age: 45–65 years Gender: 1,512 males and 1,848 females Smoking status: recorded Systemic health: patients completed a health questionnaire	-Full mouth examination (6 sites per tooth) -The severity of PPD and CAL was classified according to the term “clinically-established periodontitis” and was given by two measures: number or percentage of one diseased site, with a PPD of ≥5 mm and CAL of ≥6 mm in two or more teeth	Diagnosis of HT as per patient self-reported questionnaire	For PPD>5mm= -Unadjusted OR=0.192, 95% CI (0.12–0.31) p=0.000 -Adjusted OR= 1.268, 95% CI (0.85–1.51) p=0.000 For CAL>6mm= -Unadjusted OR= 0.039, 95% CI (0.02–0.09) p=0.000 -Adjusted age, sex, smoking: OR= 1.472, 95% CI (0.58–3.76) p=0.000 Regression analysis considers periodontal disease as dependent variable!	The findings of the current research confirm the results of previous investigations showing that a number of systemic medical conditions are significantly associated with PPD or CAL. Significant associations were also observed between increased PPD/CAL and the occurrence of HT
D’Aiuto *et al.,* (2008) US Association of the Metabolic Syndrome with Severe Periodontitis in a Large U.S. Population-Based Survey	N=13,677 individuals. Age & Gender: As per groups -No periodontitis (N=11,758), age 38.7(37.8 –39.5), 51.2% females -Moderate periodontitis (N=1,582), age 54.2(53.0 – 55.5), 40.0% females -Severe periodontitis (N=337), age 52.3(49.8–55.3), 31.7% females Smoking: recorded Systemic health: Data obtained from (NHANES III) conducted in 1988–1994 on a national probability sample of non-institutionalized, non-military American population	Page & Eke, (2007)	High BP (SBP: 130 mm Hg or DBP: 85 mm Hg or on BP medication)	-Moderate and severe periodontitis: higher prevalence of HT (51–56%, 95% CI 47– 64 *vs.* 27%, 95% CI 25–29) (all *p*=0.001), compared with those with no or mild periodontitis. -A 10% increase in the periodontal pocket extent with an OR of 1.13, 95% CI (1.03–1.24), p=0.05 for metabolic syndrome. Moreover, there was a positive association of each periodontal marker with each individual component of the metabolic syndrome (i.e. hypertension OR was approximately 1.09 according to fig.1) even after adjusting for confounders.	Severe periodontitis is associated with metabolic syndrome in middle-aged individuals. Further studies are required to test whether improvements in oral health lead to reductions in cardiometabolic traits and the risk of metabolic syndrome or vice versa

Author, year, country, title	Population of study	Periodontal criteria	Blood pressure (BP)	Observed effect (expressed as OR, RR, etc…)	Publication conclusions
Desvarieux *et al.,* (2010) US Periodontal bacteria and hypertension: the oral infections and vascular disease epidemiology study (INVEST)	N= 653 patients Age ≥55 years old. Males were younger than female (67 ± 8 vs. 70 ± 9 years, p<0.001) Gender: 60% females. Systemic health: MH obtained through interview using standardized questions adapted from the Centers for Disease Control and Prevention Behavioral Risk Factor Surveillance System	6 points PPD chart with a UNC15 probe + up to 8 subgingival plaque samples form the 2 most posterior teeth -Periodontitis defined as per: 1. based on DC/AAP guidelines (Page &Eke, 2007): mild, moderate, severe. 2. Based on previous data from INVEST (Demmer *et al* 2008): Periodontitis= % PD≥3mm	BP and hypertension: 1. SBP ≥140 mmHg, DBP≥90 mmHg or 2. Taking antihypertensive medication, or 3. Self-reported history 2 BP measurements separated by 15 min Biomarkers: CRP and WBC collected	A) HT prevalence, (Demmer definition): mean SBP or mean DBP across tertiles of % PD≥3: 57%, 59% and 70% (p=0.004); 136, 138, 143 mm Hg (p= <0.0001); and 77, 77, 82 mmHg (p <0.0001), respectively. B) The prevalence of HT among participants defined as "healthy" or having either moderate or severe periodontitis was 72%, 58% and 66% (p for linear trend=0.64), respectively The OR of HT was 3.05, 95%CI [1.60-5.82] times greater among participants in the third vs. first tertile of (bacterial) etiologic burden; after further adjustment for WBC and CRP, the OR increased to 3.93, 95%CI [1.76- 8.76]	This data provided evidence of a direct relationship between the levels of subgingival periodontal bacteria and both SBP, DBP as well as HT prevalence
Franek *et al.,* (2009) Poland Association of chronic periodontitis with left ventricular mass and central blood pressure in treated patients with essential hypertension	Periodontitis group (CPITN 3–4), N= 50 patients Gender: 23 men. Age= 51.4 ± 5.2 years, Non-periodontal cases (CPITN 1-2), N=49 patients, 20 men, 49.3 ± 5.5 years. Smoking status and medical history: recorded including history of hypertension, medication, and nicotine use	CPITN Severe perio (3-4 CPITN) Non-moderate perio (1-2 CPITN)	-Office BP was recorded -Aortic BP and pulse wave velocity (PWV) were measured by a non-invasive automated device (Sphygmocor, Atcor Medical)	CPITN 1-2 vs CPITN 3-4: SBP= 131±11 vs 128±9mm Hg, p=0.29; DBP= 85±10 vs 82 ± 7 mm Hg, p=0.12 Aortic SBP: 116±15 v 124±17mm Hg, p<0.05; Aortic DBP: 78±9 v 79±10 mm Hg, p=0.56 Partial regression coefficient: 4.01, CI (0.27- 7.76), p=0.03: an association has been found with the duration of HT and CPITN	More severe forms of periodontitis are associated with increased CBP and LVM in patients with primary HT

Author, year, country, title	Population of study	Periodontal criteria	Blood pressure (BP)	Observed effect (expressed as OR, RR, etc…)	Publication conclusions
Franek *et al.,* (2010) Poland Blood pressure and left ventricular mass in subjects with type 2 diabetes and gingivitis or chronic periodontitis	N= 155 patients with Diabetes Age: mean age 61.1 ± 6.9 years, Gender: 67 F, 88 M Smoking: Excluded smokers Systemic health: Diabetes II was an inclusion criteria	Full mouth exam (4 sites PPD) according to Offenbacher *et al* 2007 (5 sub-classifications): 1.BGI-H= PD≤3mm and BOP extent score <10% 2.BGI-G= PD≤3mm and BOP extent score >10% 3. PI=PD≥4 mm with BOP≤10% 4. P2= PD≥4 mm with BOP ≥10%≤50% 5. P3= PD≥4 mm with BOP ≥50%	1- Office blood pressure 2-Central blood pressure	-Systemic BP (SBP/DBP): Healthy (H): 130±14/ 79±8 Gingivitis (G): 136±12/ 79±7 Periodontitis (PI): 140±11/ 84±7 Differences statistically significant for SBP but not for DBP. -Central BP (SBP/DBP): Healthy: 116.2 [106.5-125.9]/ 76.1 [70.9- 81.3] Gingivitis: 124.2 [121-127.3]/ 79.9 [78.2-81.6] Periodontitis (PI): 131.8 [125.7- 137.9]/ 84.6 [81.3- 87.8] Differences statistically significant for SBP and DBP.	In subjects with type 2 diabetes, periodontitis and gingivitis are associated with increased LVM and periodontitis is associated with increased central and systemic BP
Fukui *et al.,* (2012) Japan Periodontal Status and Metabolic Syndrome in Middle-Aged Japanese	N=6,421patients Age: 34-77 years Gender: Different age range reported Smoking status: smoking habit, as a categorical variable was used in statistical analyses: 1) never smoker; 2) former smoker; 3) current light smoker (<20 pack-years); and 4) current heavy smoker (≥20 pack-years) Systemic health: Each individual completed a self-administered questionnaire in advance that included their lifestyle habits and systemic disease treatment status	Periodontal condition based on the method of the Third National Health and Nutrition Examination Survey was examined (Brown et al. 1996)	High BP (≥130 mmHg/85 mmHg)	Periodontal status was significantly associated with each component of MetS, and MetS itself (i.e. the percentage of patients with SBP≥130 or DBP ≥85 was 34.6% and 28.0% for moderate/severe vs non/mild periodontitis respectively (p=<0.0001) According to CAL, was 32.1% vs 27.9% for moderate/severe vs non/mild periodontitis respectively (p=<0.0001)	The results of this study suggest that periodontal status, particularly in individuals suspected to have untreated periodontal infection indicated by ≥4 mm PD, is significantly associated with MetS

Author, year, country, title	Population of study	Periodontal criteria	Blood pressure (BP)	Observed effect (expressed as OR, RR, etc…)	Publication conclusions
Furuichi *et al.,* (2003) Japan Associations of periodontal status with general health conditions and serum antibody titers for *Porphyromonas gingivalis* and *Actinobacillus actinomycetemcomitans*	N=1,314 Age >40 years, Gender: 746 females, 568 males Smoking status: measured by questionnaires (pack-years) Systemic health: collected by a team of medical doctors	CPITN, IgG antibodies titers for Pg and Aa	Measured	Mean SBP (mm Hg): CPITN: 134 ± 21.6 CPTIN 3: 132.1 ± 21.5 CPITN0/1/2: 132.2 ± 22.8 No statistically significant differences among groups Mean DBP (mmHg): CPITN 4: 78.8 ± 10.8 CPTIN 3: 77 ± 11.2 CPITN0/1/2: 77.4 ±12 No statistically significant differences among groups Simple regression: OR of HT→ perio: SBP:1.07 [0.94-1.23), p=0.3003 DBP:1.19 [1.04-1.37], p=0.011 Multiple regression OR of HT→ perio: SBP: 0.86 [0.70-1.05], p=0.144 DBP: 1.27 [1.05-1.55], p=0.0167	Significant associations between periodontal status and several health conditions were found in the adult population examined, including gender, smoking habit, diastolic blood pressure, white blood cell counts, CRP, and serum IgG antibodies to *P. gingivalis* fimbriae, IgG P. gingivalis whole cell, and IgG *A. actinomycetemcomitans* whole cell titers
Gomes-Fihlo *et al.,* (2016) Brazil Severity of periodontitis and metabolic syndrome: is there an association?	N= 419 patients from the Diabetes and hypertensive treatment center. Mean age: 59 ±13.3 years [24-89] Gender: 259 females Smoking: questionnaire Systemic health: Questionnaire and clinical and oral examinations and lab test done	Full mouth periodontal charting. Periodontitis: ≥4 teeth with ≥1 site with a PD ≥4 mm, CAL ≥3 mm, and BOP Severity as per Page & Eke, (2007)	Measured with stethoscope SBP≥130 mmHg DBP≥85 mmHg	Periodontitis and MetS adjusted OR= 2.11, 95% CI (1.01-4.40), p= 0.05 Adjustment for sex, age, household density, alcoholic beverage consumption, smoking habit, and CVD 62.80% (91 patients) with periodontitis had HT versus 67.60% (184 patients) without periodontitis had HT p=0.32 70.30% (102) patients with periodontitis had SBP≥130 vs 59.10% (162 patients) with non-periodontitis p=0.02 and 49.70% (72 patients) with periodontitis had DBP≥85 vs 43.10% (118 patients) without periodontitis	The results suggest that periodontitis is associated with MetS, and that MetS prevalence is related to severe periodontitis

Author, year, country, title	Population of study	Periodontal criteria	Blood pressure (BP)	Observed effect (expressed as OR, RR, etc…)	Publication conclusions
Gordon *et al.* (2018) The US Association of clinical measures of periodontal disease with blood pressure and hypertension among postmenopausal women	N: 1341 postmenopausal women Age: 66.7 (7.0) Smoking status: Reported as never, former or current. Systemic health: Assessed by questionnaire as part of WHI-OS or OsteoPerio study, Height and weight measured, BMI, medication via inventory	Full mouth charting. PPD with Florida probe, CAL assessed with manual probe and bone loss with radiographs evaluation. CDC-AAP (Page & Eke, 2007) ACH loss (Alveolar crestal Height)	BP measured twice on the same day. Hypertension was also defined if patient answered yes to HTN dx in questionnaire or was taking antihypertensive medication	16.2% of patients with severe periodontitis had hypertension versus 26.2% with none or mild hypertension had Hypertension 28.6% of patients with severe alveolar bone high loss had HT versus 23.1% of patients with no alveolar crestal height loss Alveolar crestal height (ACH) and gingival bleeding on probing were associated with higher SBP in crude but not multivariable adjusted models. Neither probing pocket depth (PPD) nor severity categories of periodontitis were associated with SBP: OR moderate perio→HT= 1.06 CI(0.80-1.40) *p.*0.59 OR severe perio→HT= 1.03 CI(0.71-1.50) *p.* 0.88	These results suggest that measures of oral health including CAL and number of teeth missing are associated with blood pressure in postmenopausal women.
Gorska *et al.,* (2017) Poland Correlation between the state of periodontal tissues and selected risk factors for periodontitis and myocardial infarction	N=417 patients hospitalized due to recent MI. Age: 25 - 69 years. Gender: 92 women Smoking status: current (smoking ≥10 cigarettes/day continuously for at least 5 years), smoking in the past and never; Systemic disease: checked by interview and recorded	CPI index	Arterial HT was defined as SBP ≥ 140 mm Hg or DBP ≥ 90 mm Hg or use of antihypertensive drugs.	The percentage of pockets of PPD ≥ 4mm was 29.3% ± 25.4 for people with HT vs 23.4% ± 21.9 for people without HT. Similarly, the mean PD was 3 ±1.1 for HT vs 2.8± 0.9 for non-HT	The degree of severity of periodontal disease can impact hypertension and diabetes, which could potentially influence the occurrence and course of CVD

Author, year, country, title	Population of study	Periodontal criteria	Blood pressure (BP)	Observed effect (expressed as OR, RR, etc…)	Publication conclusions
Han *et al.* (2018) Korea Clinical implication of fasting glucose and systolic/diastolic blood pressure on the prevalence of periodontitis in non-diabetic and non-hypertensive adults using nationally representative data	N: 8,341 respondents without diabetes and hypertension of which (1968 had periodontitis) Age: >19 years old Periodontitis patients: 56.24 (1.17) years old No periodontitis patients: 44.65 (0.66) years old Smoking status: classified as current smokers and non-smokers Systemic health: Heath status of each participant was evaluated and measured.	Community Periodontal Index (CPI) (WHO) Periodontitis: CPI> 3 Moderate periodontitis= CPI 3 Severe periodontitis= CPI 4	BP measured using sphygmomanometer (Baumanometer; W.A. Baum Co., Inc., Copiague, NY, USA). SBP and DBP were performed two times with a 5-min interval, and the average of the two measurements was used for the analysis. DX=SBP/DBP ≥140/90 mmHg or taking anti-HT	-The mean blood pressure in perio vs non-perio was: SBP= 112.91±0.31 for perio patients versus 109.73±0.18 for non-perio; p<0.001 DBP=74.01±0.22 for perio patients versus 91.66±0.15 for non-perio patients; p<0.001 -Most adjusted OR: The ORs and 95% CIs for SBP of 90≤x<100= 1.116 (0.591-2.107) 100≤x<110= 1.165 (0.624-2.175) 110≤x<120= 1.238 (0.673-2.278) 120≤x<130= 1.008 (0.538-1.888) 130≤x<140= 1.042 (0.545-1.993) Adjusted for: Model 2 + number of natural teeth, frequency of tooth brushing per day, use of secondary oral products and duration of sleep adjusted.	The association between fasting glucose/blood pressure and periodontitis was proven by multiple logistic regression analyses after adjusting for confounding factors among non-diabetic and non-hypertensive Korean adults.
Han *et al.,* (2010) Korea The association of metabolic syndrome with periodontal disease is confounded by age and smoking in a Korean population: the Shiwha–Banwol Environmental Health Study	N=1,046 Age: 42.3±12.2 (18 – 84) years. Gender: 457 men Smoking: Measured: over 20 packs in lifetime (yes/no) Systemic health assessment questionnaire	CPI (WHO) Periodontitis (CPI 3 or CPI 4)	HT (≥130/85 mm Hg or on blood pressure medication).	48.7% (131 patients) with CPI 3-4) had HT versus 51.3% (138 patients) with CPI 2-1 and HT MS was strongly associated with periodontitis (OR= 1.7, 95% CI= [1.22–2.37], p=0.002	Our results suggested that MS might be associated with periodontitis and the association was confounded by age, gender, and smoking. MS with high glucose and hypertension showed the higher impact on this link

Author, year, country, title	Population of study	Periodontal criteria	Blood pressure (BP)	Observed effect (expressed as OR, RR, etc…)	Publication conclusions
Han *et al.,* (2016) Korea Excessive consumption of green tea as a risk factor for periodontal disease among Korean Adults	N=16,726 Age/gender: 40.57 ± 0.26/male 45.7% (0.5) (non-perio) Age/gender: 52.08 ± 0.31/male 59.4% (0.7) (perio) Smoking status: was defined according to self-reported cigarette use and based on current smoking habits Systemic health: Questionnaire and clinical examination	CPI on 10 specific index teeth. A CPI of 3 or 4 was considered as having periodontitis (moderate and severe periodontitis), respectively	SBP ≥ 140 mm Hg or DBP ≥ 90 mmHg or taking hypertensive medication	38.9% ± 0.9 of the patients with periodontitis had HT versus 20.8% ± 0.5 of the non-periodontal patients had HT p<0.0001	In conclusion, excessive consumption of green tea may be considered as a risk factor for periodontal disease among Korean adult
Holmlund *et al.,* (2006) Sweden Severity of periodontal disease and number of remaining teeth are related to the prevalence of myocardial infarction and hypertension in a study based on 4,254 subjects	N= 4,254 (3,352 patients referred to Gavle University perio department + 902 of general population) Periodontal severity index (PDSI): PDSI<1 (no disease): N = 510; PDSI 1-1.9 (mild): N = 201; PDSI 2-2.9 (moderate); N = 281; PDSI >2.9 (severe) N = 726 Mean age: 53 ±14 years Gender: 2,388 females Systemic health and smoking: questionnaire about their medical history, medications, and smoking habits	Full-mouth probing (4 points) PPD ≥5 mm was regarded as diseased. Full-mouth radiographs Periodontal severity index (PDSI) was calculated	Self-reported questionnaire. HT was defined as drug treatment for HT	Relationship between the prevalence of HT and periodontal bone loss: (OR= 1.32, 95% CI [1.13–1.54], p<000.5) Relationship between number of diseased periodontal pockets and HT: OR= 1.01, 95% CI [1.01–1.02], p<0.0001	Severity of periodontal disease was related to HT independent of age but to the prevalence of MI in middle-aged subjects only. Number of diseased pockets was significantly for HT only. Number of teeth associated with prevalence of MI independent of age (not HT). These data support the view that oral health is related to cardiovascular disease in a dose-dependent manner

Author, year, country, title	Population of study	Periodontal criteria	Blood pressure (BP)	Observed effect (expressed as OR, RR, etc…)	Publication conclusions
Iwashima *et al* (2014) Japan Additive interaction of oral health disorders on risk of hypertension in a Japanese urban population: the Suita Study	N= 1,643 participants Mean age= 66.6 ± 7.9 y Gender: 43.4% of men Smoking status: recorded Systemic health: questionnaire on individual personal habits and present illnesses	A modified CPITN (in 8 designated molars; first and second molars and 2 incisors) Gingival bleeding Occlusal support or masticatory performance was recorded by means of the Eichner index	Hypertension: SBP ≥140mm Hg and/or DBP ≥90 mm Hg, or use of antihypertensive medication	-Significantly worse CPITN in HT subjects, higher prevalence of gingival bleeding, lower tooth number, and worse Eichner index. After adjustment, no individual oral health markers were significantly associated with HT. -CPITN4 OR for HT= 1.05, CI 95% [0.96-1.16], p=0.24 adjusted for multivariable- model included age, sex, body mass index, diabetes, dyslipidaemia, eGFR, smoking -CPITN4 and BoP+ OR for HT= 1.72 [1.17-2.50], p<0.01 adjusted for multivariable-model included age, sex, body mass index, diabetes, smoking, dyslipidaemia, eGFR	There is an additive relationship between oral health disorders and risk of HT. Our results suggest that the existence of moderate or severe oral health disorders—that is, several concomitant oral health disorders—is associated with risk of HT
Khocht *et al.,* (2017) USA Gingival fluid inflammatory biomarkers and hypertension in African Americans	N=21 subjects with HT and 26 non-HT group Mean age= 52.3 ±1.7 in non-HT group vs 52 ±1.9 in HT group Gender: 30.8% of males in non-HT group vs 23.8 % of males in HT group Smoking status: no cigarette smoking at least 6 months prior to study Systemic health: free from other systemic illness. A comprehensive medical history interview was undertaken	Full mouth assessment. Definition: Eke *et al*. 2012 Gingival crevicular fluid (GCF) was collected non-diseased sites and in diseased sites as having PD>4 mm with equivalent or greater CAL	BP (Chobanian *et al,* 2003) HT: self-reported, BP between 140/90 and 160/100 mm Hg GCF for: 8-isoprostane, IL-1B, monocyte chemoattractant, protein 1 (MCP-1), *TNFα* *CRP, MMP8*	-Regression analysis (adjusted for demographics, smoking, BMI, and plaque index) showed almost twice as much AL in hypertensive (HTN= 0.87±0.13 as non-HTN subjects 0.49±0.11). -Biomarker levels and TNFα rates as a function of HT and PD. Regression analysis (transformed data) showed higher rates of TNFα and higher levels of all other biomarkers in subjects with deep pockets (all p< 0.05). -In subjects with shallow pockets, higher levels of CRP were found in the GCF of HT people than normotensive subjects (p= 0.02). -Other biomarkers showed no statistically significant associations with HTN; however, their levels were significantly higher in subjects with deep pockets	In conclusion, individuals with elevated blood pressure show heightened levels of 2 inflammatory mediators in the gingival fluid collected from non-diseased sites, which may exacerbate periodontal tissue damage

Author, year, country, title	Population of study	Periodontal criteria	Blood pressure (BP)	Observed effect (expressed as OR, RR, etc…)	Publication conclusions
Koo et al. (2018) Korea Prevalence and Risk Factors for Periodontitis Among Patients with Metabolic Syndrome	N: 13196 Periodontitis group: 6598; mean age= 57.2±13.0 years old and 53% males Non-periodontitis group: 6598; mean age= 57.4±13.0 years old and 51% males Smoking: Smoker= current smoker or >100 cigarettes in their whole life Systemic health: Assessed with questionnaire and clinical measurements.	CPI (WHO) Periodontitis CPI=3/4	SBP/DBP= ≥140/90 mmHg or taking antihypertensive medication	-Prevalence: Perio group had 43% of patients with hypertension) versus non-perio group 40%; p <0.001 -Mean BP : Perio group: SBP = 123.5±17.3 mm Hg DBP= 77.1±10.5 mm Hg Non-perio: SBP= 122.3±16.8 mm Hg p<0.001 DBP= 76.8±10.2 mm Hg p=0.150 -OR Metabolic syndrome→ Perio: OR=1.14 (95%CI: 1.03-1.27) with 1 component OR=1.52 (95%CI: 1.13-2.05) with 5 components (adjusted for sex, educational level, BMI, smoking, FPG level)	Patients with Met Syndrome had 1.12-fold increase higher risk of periodontitis than those without. The risk of periodontitis increased as the number of MS components increased.
Kushiyama *et al.,* (2009) Japan Relationship between metabolic syndrome and periodontal disease in Japanese adults	N= 1,070 subjects Age > 40 years old Gender: 281 males Smoking status: former or current smoker Systemic disease: self-administered questionnaire	CPI (WHO) Divided by (0-3) or 4 scores (periodontitis)	HT: BP≥130/ 85 mm Hg	188/316 patients with CPI=4 vs 325/754 patients with CPI code of 1-3 had HT OR of HT→Perio: -Crude: OR=1.94 [1.49-2.53], p<0.001 -Adjusted for age, gender, and smoking habits: OR=1.59 [1.20-2.11], p= 0.001	This study supports the suspected but unproved relationship between metabolic syndrome and periodontal disease

Author, year, country, title	Population of study	Periodontal criteria	Blood pressure (BP)	Observed effect (expressed as OR, RR, etc…)	Publication conclusions
Kwon *et al.,* (2011) Korea The relationship between periodontitis and metabolic syndrome among a Korean nationally representative sample of adults	N= 7,178 subjects Age: >19 years old Gender: 2694 males Smoking status: past, current or non-smoker Systemic disease: self-administered questionnaire	CPI (WHO) Divided by (0-3) or 4 scores	HT: BP≥ 130/85 mm Hg	OR HT→Perio= 1.12 [0.94–1.34] -Periodontitis patient had mean value of SBP= 120.12 [119.11– 121.14] and DBP= 76.94 [76.29– 77.58] vs -Non-periodontitis patients: SBP= 110.84 [110.15– 111.53] and DBP= 73.61 [73.07– 74.16] Prevalence: 1,167 patients (36.2% [33.9–38.6]) of periodontitis patients vs 718 (18.5% [17.1–20.1]) of non-periodontitis had HT	MS is associated with periodontitis.
Lysek *et al.,* (2015) Poland Association between central and peripheral blood pressure and periodontal disease in patients with a history of myocardial infarction	N= 99 patients (patients had had MI 6-18 months prior to study) Mean age= 60.5 ± 8.71 years. Gender: 71 men Smoking status: considered smokers if they reported smoking at least cigarette during the month preceding the interview or had at least 10 ppm of carbon monoxide in exhaled air. Systemic health: data on the history of ischemic heart disease, drug use, demographic characteristics, and the presence of risk factors were collected by an interview using a standard questionnaire	CPI	HT was defined as peripheral BP ≥140/90 mm Hg or higher and central BP≥130/90 mm Hg, as per Cheng *et al* (2013) All participants took at least 1 hypertensive drug	-Central and peripheral BP increased with an increase of the CPI -After adjustment for covariates: Patients from the CPI 3+4 group were found to have almost 3 times higher odds of central BP of 130/90 mm Hg or higher OR=2.90, 95% CI [1.14–7.66] and more than 3 times higher odds of peripheral BP of 140/90 mmHg or higher OR 3.38, 95% CI [1.30–9.42] vs -Patients from the CPI 1+2 group. 58.3% of people with CPI=3+4 had peripheral BP ≥140/90 mmHg versus 25.5% of people with CPI=1+2 p<0.001 also 62% of people with CPI=3+4 had central BP ≥130/90 mmHg versus 31.4% of people with CPI=1+2 p=0.002 -CPITN=3/4 had mean pSBP=144.8 ± 21.68, cSBP=130.3 ± 20.55, pDBP=87.4 ± 12.48, cDBP=92.3 ± 12.49 vs CPITN 1+2 pSBP=129.0±19.14, cSBP=116.3±17.19, pDBP=78.8 ± 10.29, cDBP=80.6 ±11.01 p<0.001	In conclusion, there is a similar association be-tween PD and central and peripheral BP. The association between PD and BP may partially ex-plain the CV risk related to chronic PD. High antibody titer against *P.g.* not related to central or peripheral BP

Author, year, country, title	Population of study	Periodontal criteria	Blood pressure (BP)	Observed effect (expressed as OR, RR, etc…)	Publication conclusions
Machida *et al*., (2014) Japan Severe Periodontitis Is Inversely Associated with Coffee Consumption in the Maintenance Phase of Periodontal Treatment	N= 430 patients with chronic periodontitis in the maintenance phase Mean age=66.4 ± 9.9y Gender: Males: 86 (20.8%); Females: 328 (79.2%) Smoking status: current, never and former. Systemic health: not mentioned how it was recorded, but data of diabetes, hypertension, dyslipidaemia and heart disease given	Chronic periodontitis was defined as ≥1 tooth site with PPD ≥4 mm 6 points pocket chart recorded Periodontitis severity according to Page&Eke (2007)	Not mentioned diagnosis or how it was recorded	The presence of moderate/severe periodontitis was correlated with presence of HT (Adjusted OR = 1.99, 95% CI (1.07-3.71), $p< 0.05$. With severe periodontitis: Adjusted OR = 2.46, 95% CI (0.82-2.59), p= 0.196 84 of moderate-severe periodontitis patients (28.7%) had HT vs 17 of non-periodontitis or mild periodontitis (14.0%) had HT p=0.002	There appears to be an inverse association between coffee consumption (≥1 cup/day) and prevalence of severe periodontitis in the maintenance phase of periodontal treatment
Moghadam *et al.,* (2016) Iran A Relationship between Tooth Loss and Periodontal Disease with Increased Blood Pressure in Adults: A Population-Based Study in Iran	N= 700 adults Mean age=45.62 ±7.4 Gender: 46.7% female vs 53.3% male Smoking status: the participants were categorized into five groups with respect to smoking (Volzke *et al*. 2007) Systemic health: Systemic diseases were investigated (Questionnaire)	Periodontal chart was completed for each person by one examiner. The number of natural teeth in both jaws was counted for each person. Patients were categorized in four groups: periodontitis, gingivitis, healthy, complete edentulism	BP measured in clinical examination under standardised conditions (Stethoscope) BP≥140/90 mmHg	A positive and significant correlation was also observed between tooth loss and SBP Periodontal disease and BP: per unit change in periodontal disease from Class 1 to Class 4, SBP increased 4.95mm Hg (p<0.001) In addition, per unit change in periodontal disease (from Class 1 to Class 4), DBP increased 3.03mm Hg (p <0.001)	Total edentulism was in relation to increasing levels of SBP and DBP in the adult population

Author, year, country, title	Population of study	Periodontal criteria	Blood pressure (BP)	Observed effect (expressed as OR, RR, etc…)	Publication conclusions
Morita *et al.,* (2009) Japan Association between periodontal disease and metabolic syndrome	N= 2,478 employees Mean age= 43.3 years old [24-60] Gender: 2,028 men Smoking status recorded Systemic health: Blood sample and clinical examination and systemic disease recorded	(CPI) WHO Periodontitis: CPI=3/4	BP≥130/85 mmHg	43.2% (277 subjects) with periodontitis had HT vs 30% (549 subject) without periodontitis had HT. OR=1.2 (1.0-1.5), p<0.01 For HT→perio	Our findings suggest an association between periodontal disease and metabolic syndrome in Japanese workers between the ages of 20 and 60 years
Nesbitt *et al* (2010) USA Association of periodontitis and metabolic syndrome in the Baltimore Longitudinal Study of Aging	N= 190 people participating in the Baltimore Longitudinal Study of Aging. Gender: 112 men Mean age= 56.7 ± 13.3 (men) and 60 ± 12.1 (women). Smoking status: smoker/non-smoker Systemic health: Medical examination performed to ascertain the components of met syndrome	Periodontal status was determined from pantomographic images taken as part of the dental examination. Participants were scored as having: -none or slight (1–2 mm), -moderate (3–4 mm), or -severe (≥5 mm) alveolar bone loss based on radiographic assessment of crestal bone height (excluding 3rd molars)	SBP and DBP values were defined by Kortokoff phases I and V BP measured with sphygmomano-meter	-Patients with severe bone loss had higher SBP=133.7 ± 22.8 than individuals with non-slight bone loss (SBP=123.8 ± 21.9), p=0.01; and DBP=79.8 ± 14.2 vs 78.1 ± 9.6, p=0.47 -Logistic regression(periodontal disease/MetS components): -Adjusted for age, sex, race, and smoking status; participants with advanced alveolar bone loss were significantly more likely to exhibit high BP than those without periodontal disease: OR=2.12, 95% CI (0.9–4.8), *p*=0.07; -The adjusted OR for age, sex, race, smoking status and inflammation: OR= 1.97, 95% CI (0.9-4.5), p=0.1	The association of alveolar bone loss to MetS is consistent with the hypothesis that destructive periodontal disease may contribute to the development of MetS and elevations in systemic inflammation. Longitudinal studies are necessary to clarify the role of periodontal disease in the development of MetS and conditions associated with chronic inflammation

Author, year, country, title	Population of study	Periodontal criteria	Blood pressure (BP)	Observed effect (expressed as OR, RR, etc…)	Publication conclusions
Nesse *et al.,* (2010) The Netherlands Increased Prevalence of Cardiovascular and Autoimmune Diseases in Periodontitis Patients: A Cross-Sectional Study	N= 1,218 patients of which 547= 320 controls (44% males, age=33±11) + 217 periodontal patients (52% males, age=41±12) attending a dental clinic and 671 patients (32% females, ag=49±11) attending two periodontal clinics Smoking: 31%, 28%,40% in controls, dental clinic and periodontal clinic successively Systemic health: extensively validated, self-reported health questionnaire. Answers were verified by experience physician regarding the presence or absence of cardiovascular or autoimmune diseases	CPITN	From health questionnaire	Univariate unadjusted analysis: In the dental clinic and the periodontitis clinic the prevalence of HT was significantly higher in patients with periodontitis 13.4% (29 patients) compared to controls 5% (16 patients) (P <0.001) Results of logistic regression analysis: periodontitis did not predict the presence of the remaining diseases analysed in this study, OR of these diseases were not significantly increased, nor was periodontitis kept in the regression models predicting these diseases	The increased prevalence of CV and autoimmune diseases among patients with periodontitis attending dental or periodontal clinics may, at least in part, be influenced by confounding. However, the increased prevalence of DM and RA in patients with periodontitis could not be explained by confounding
Ollikainen *et al.,* (2014) Finland Association between periodontal condition and hypertension in a non-smoking population aged 30–49 years: results of the Health 2000 Survey in Finland	N= 1,296 Mean age: 40 years old Gender: 42.3% men Smoking status: only non-smokers included Systemic health: recorded as per the national survey in Finland carried out 2000-2001	When the number of teeth with deepened periodontal pockets was used as an explanatory variable in the regression models, it was categorized into five categories: 0, 1–3, 4–6, 7–11, and 12 or more teeth	The criteria for HT: BP≥140/90 mmHg or use of antihypertensive medication	-After adjusting for confounding factors, there were no consistent associations between the number or proportion of teeth with deepened (≥4 mm or ≥6 mm periodontal pockets, or the number of bleeding sextants and HT). OR (teeth with PPD ≥ 4 mm) → HT 1.03, 95% CI (1.00-1.06) (unadjusted) -OR (teeth with PPD ≥ 4 mm) → HT= 0.98, 95% CI (95-1.01) (adjusted for age, gender, educational level, body mass index, physical activity, alcohol consumption (g/week), and serum lipid composition (triglycerides, HDL, LDL) -OR (teeth with PPD ≥ 6 mm) → HT= 1.16, 95% CI (0.98-1.38) (unadjusted) -OR (teeth with PPD ≥ 6 mm) → HT= 1, 95% CI (0.97-1.04) (adjusted for same confounders as for PPD ≥ 4 mm	Periodontal pocketing and gingival bleeding did not appear to be related to HT in non-diabetic, non-smoking individuals aged 30–49 years. Further studies using experimental study designs would be required to determine the role of infectious periodontal diseases in the development or progression of HT

Author, year, country, title	Population of study	Periodontal criteria	Blood pressure (BP)	Observed effect (expressed as OR, RR, etc…)	Publication conclusions
Pietropaoli *et al.* (2018) US Poor Oral Health and Blood Pressure Control Among US Hypertensive Adults Survey 2009-2014	N: 4095 (3626 taking medication for high BP (treated HTN) and 460 were not taking medication (untreated HTN), of which 47.8% did not have periodontal disease and 52.2% had periodontal disease and. Age: reported according to no-periodontitis= 62.60 (12.94) or mild= 57.32 (12.81) /moderate= 64.17 (11.75)/ severe= 62.64 (10.53) p<0.001 Gender: reported according to no-periodontitis= (39.9% men), mild=(45.7% men), moderate= (50.5% men), severe= (62.4% men) Smoking status: current/former or never smoker Systemic health: Assessed by questionnaire, anthropologic measurements taken.	CDC/AAP (Eke et al. 2012)	BP measured by trained and calibrated physicians using a mercury sphygmomanometer according (3 readings for each patients). HTN: According to the ACC/AHA 2017 guidelines SBP/DBP>130 /80mm Hg (Whelton et al. 2017)	Mean SBP for patients with: No-perio= 131.17 (19.54) Mild perio= 128.14 (19.19) Moderate perio= 133.56 (19.14) Severe perio= 134.35(21.32) Mean DBP for patients with: No-perio= 69.58(13.88) Mild perio= 72.90(12.37) Moderate= 69.67(14.32) Severe perio 71.10(14.73) Mean SBP was about 2.3 mmHg higher in treated hypertensive adults (n=1834;133.43±19.7 mm Hg) with periodontitis than those without the disease (n=1694; 131.17±19.5 mm Hg; p<0.001). Such difference increased to about 3 mm Hg after progressive adjustment (P<0.001) Perio disease→ Risk of unsuccessful HT treatment= OR=1.19 (0.91-1.54) p=0,205 (adjusted model for age, sex, ethnicity, BMI ranges, smoking status, HbA1C, total cholesterol, HDL cholesterol, triglycerides, creatinine, education and poverty level and CRP)	A good periodontal health is associated with better SBP profile during antihypertensive therapy by about 2.3 to 3 mmHg and with lower odds of treatment failure.
Ribeiro *et al.,* (2016) Brazil Association of dental infections with systemic diseases in Brazilian native indigenous: a cross-sectional study	N= 225 indigenous Age: 19-77 years old Gender: Reported according to different analysis Smoking: information on nicotine dependence recorded. Systemic disease: structured written questionnaire (demographics, socioeconomic & health-related	-Full mouth charting -As per Figueredo *et al* (2013): Destructive PD -Severity: (Page & Eke 2007)	According to the AHA as follows: BP ≥139/89 mm Hg; or non-HT SBP <139/89 without the use of an antihypertensive	Individuals with destructive periodontal disease had a higher SBP (124± 20.34 mm Hg) than those without destructive periodontal disease (117.52 ±16.54 mm Hg, p= 0.01). DBP =76.90 ± 14.99 for perio vs 75.1 ± 15.89 for non-perio, p=0.39 51.9% of patients with destructive periodontal disease had HT vs 48.1% of patients with no destructive periodontal disease	Dental infections were found to be associated with HT in the present population. Thus, patients diagnosed with HT should be referred for dental evaluation and vice versa

Author, year, country, title	Population of study	Periodontal criteria	Blood pressure (BP)	Observed effect (expressed as OR, RR, etc…)	Publication conclusions
Rivas-Tumanyan *et al.,* (2013) Puerto Rico Periodontal disease, hypertension and blood pressure among older adults in Puerto Rico	N= 182 subjects from the Puerto Rican Elderly Dental Health Study (PREDHS) from an on-going representative cohort of Puerto Rican Elderly (PREHCO). Age:>70 years old Gender: men Smoking status: from the responses to the PREHCO study interview Systemic health: in-person interview on current oral health status, history of dental diseases and procedures	Page & Eke (2007) severe periodontitis (≥2 teeth with AL ≥6mm and ≥1 tooth with PD ≥5mm) By dental examiners performing home visits	BP ≥140/90 mmHg Stages according to JNC7 report Stage 0 if SBP<140 and DBP<90; Stage 1 if 140 ≤ SBP<160 or 90 ≤DBP < 100; Stage 2 if SBP≥160 or DBP≥ 100) By dental examiners receiving training and calibration	Several OR given according to different models and adjustments: 1-No association between severe periodontal disease and history of HT diagnosis (OR=0.99, 95% CI (0.40–2.48) 2- Participants with severe periodontal disease had 2.93 times higher odds of having HT upon examination (multivariate-adjusted OR=2.93, 95% CI (1.25–6.84). The OR estimate for severe periodontal disease remained strong and statistically significant after additional adjustment for anti-hypertensive medications; OR= 3.00, 95% CI (1.28–7.03) or for number of teeth (model 4 OR=2.87, 95%CI (1.22–6.74) 3- 64% of the patients with severe periodontal disease had HT versus 70% of patients without severe periodontitis. Additionally, 75% of patients with severe periodontitis presented with high BP vs 55% of those without severe periodontitis	The results suggest that periodontitis may contribute to poor blood pressure control among older adults
Shamsuddin *et al.,* (2015) Malaysia Dental records taken retrospectively for the cross-sectional study. Sample size issues!	N= 90 patients with chronic periodontitis. Age: 41-61 years old Gender: 56 males (62.2%) and 34 females (37.8%) Smoking status: 80% non-smokers according to records. Systemic health: demographic and clinical from patients' record	Severity of CP was classified according to the (CAL) as mild CP if CAL=1-2 mm, moderate CP if CAL=3-4 mm and severe CP if CAL≥5 mm or more (Flemming, 1999)	As recorded in medical notes. underreported medical problems	-The prevalence of HT in chronic periodontitis patients was 12.2% -No significant association between HT and severity of CP (p=0.229). -However, OR as reported by Martin Cabezas et al. 2016; OR=7.24, 95% CI (1.6-38.36)	In conclusion, the prevalence of HT in chronic periodontitis patients was 12.2%. There was no significant association between HT and severity of CP (p=0.229)

Author, year, country, title	Population of study	Periodontal criteria	Blood pressure (BP)	Observed effect (expressed as OR, RR, etc…)	Publication conclusions
Shimazaki *et al*. (2007) Japan Relationship of metabolic syndrome to periodontal disease in Japanese women: The Hisayama Study	N= 584 patients Age:40-79 y Gender: All women Smoking status: current or past smoker Systemic health: self-administered questionnaire in advance that included a medical history of diabetes, hypertension, smoking, and medication use; the questionnaire was checked by trained nurses	Mesio-buccal and mid- buccal sites for all of the teeth in 2 quadrants randomly selected, one maxillary and one mandibular Divided in 2 groups: < 2.0 mm (N = 484, 82.9% of all participants) and ≥2.0 mm (N= 100, 17.1%) And also divided by CAL: <3.0 mm (N= 547), 93.7% and ≥ 3.0 mm (N = 37), 6.3%	Measured and HT: BP>130/85 mm Hg	-Mean SBP (CAL) CAL<3mm= 127.2 ± 20.6 mm Hg CAL≥3 mm= 131.1±22.8mm Hg -Mean DBP (CAL): CAL<3mm= 76.4 ± 10.4 mm Hg CAL≥3 mm= 76.1 ± 10.4 mm Hg -Prevalence according to PPD: 53% of patients with PPD ≥2 mm had HT vs 41.9% with PPD<2mm OR: HT→perio Adjusted (age and smoking): OR=1.5, 95% CI (0.9-2.3) not statistically significant OR: HT→ perio Unadjusted: OR=1.6, 95% CI (1.0-2.4), p<0.05 -Prevalence according to CAL:56.8% (21 patients) with CAL3≥mm had HT vs 43% (235 patients) with CAL<3 had HT OR: HT→perio Adjusted (age and smoking): OR=1.3, 95% CI (0.6-2.7) not statistically significant OR: HT→ perio Unadjusted: OR=1.7 (0.9-3.4) not statistically significant	These results indicate that MetS increases risk of periodontitis and suggest that people exhibiting several components of MetS should be encouraged to undergo a periodontal examination

Author, year, country, title	Population of study	Periodontal criteria	Blood pressure (BP)	Observed effect (expressed as OR, RR, etc…)	Publication conclusions
Thanakun *et al*., (2014) Thailand Association of untreated metabolic syndrome with moderate to severe periodontitis in Thai population.	N=125 participants Age: mean age (years)= 47.0 [42.0, 56.0] Gender: 53 (42.4%) males, 72 (57.6%) females Smoking status: measured (never, former, current) Systemic health: medical records and clinical assessment for glucose, HDL-C, waist circumference, etc.	-Full mouth assessment (6 sites) -Periodontitis according to AAP: CAL ≥3 mm (moderate to severe periodontitis) Savage *et al* (2009): PPD ≥4 mm	Measured BP>130/85 mm Hg	-Mean (SBP/DBP), According to PPD: None/mild periodontitis: 120 (112-130)/78 (72- 83); p=0.001 Moderate/severe periodontitis: 133 (118.8-140.3)/85.5 (78-91.3); p<0.0001 -Mean (SBP/DBP): According to CAL: None/ mild periodontitis: 120 (111-130); 78 (73-82), p=0.04 Moderate/ severe periodontitis: 128 (117-139.3/82 (73- 89), p=0.04 -SBP→moderate/severe perio: OR= 4.86, 95%CI (0.54-43.44) p=0.16 -DBP→moderate/severe perio OR= 53.22, 95%CI (3.22-89.79) p=0.01	There may be a relationship between untreated MetS and periodontitis in Thai people. Periodontal diagnosis should be regularly conducted in patients with MetS
Tsakos et *al*., (2010) US Is periodontal inflammation associated with raised blood pressure? Evidence from a National US survey.	N= 13,994 Age: >17 years old Gender: 6617 men Smoking status: 1) Current smokers (reporting current smoking of cigarettes, cigars and/or pipe); 2) Non-smokers; 3) Non-respondents. Systemic health: Collected data in relation to health status and diseases and possible confounders	-Page *et al* (2007) case definition of periodontitis -Also continues periodontal measures on the extent and severity of the disease	Presence of mean SBP at least 140 mmHg and/or DBP at least 90 mmHg and also including participants on antihypertensive medication	For a 10% greater extent of gingival bleeding, the average SBP was higher by 0.5 (0.3-0.6) mmHg. An OR of 1.1. for gingival bleeding and an OR 1.2NS (0.7–2.1) were obtained for diagnosis of HT after full adjustment. Dose response: individuals with moderate and those with severe periodontitis had higher SBP by 10.7 (9-12.4) and 11.7 (8.6-14.7) mmHg, respectively, than individuals without or with mild periodontitis	Gingival bleeding, a marker of current periodontal inflammation, was consistently and significantly associated with raised SBP and an increased odd of hypertension in the US adult population, even after adjusting for the effect of socio-demographic, behavioural, and physiological factors and chronic diseases

Author, year, country, title	Population of study	Periodontal criteria	Blood pressure (BP)	Observed effect (expressed as OR, RR, etc…)	Publication conclusions
Tu *et al.,* (2013) Taiwan Relationship between metabolic syndrome and diagnoses of periodontal diseases among participants in a large Taiwanese cohort.	N= 33,740 Age: Gender: 18,469 women and 15,271 men. Smoking status: former or non-smoker Systemic health: check up by GP and recorded	A tooth was diagnosed with periodontal diseases if presented with a combination of clinical signs, such as tooth mobility, gingival inflammation and periodontal pocketing Periodontitis group: people with at least one tooth with diagnosis of periodontitis	-BP ≥130/85 mmHg or treatment for hypertension	Female participants in the periodontitis group had higher BP after adjusting for confounders. Male participants in the periodontitis group exhibited higher waist circumference, fasting glucose, postprandial glucose, HbA1c, TG and CRP level compared to the reference group and independent. Periodontitis group in women showed greater odds of HT: OR=1.33, 95%CI (1.23-1.43), p<0.001, than patients with gingivitis, whereas men showed an OR=1.009 (0.934–1.09), p= 0.813	A small but statistically significant association between MetS and the diagnosis of periodontal diseases was found in Taiwanese women and a weaker association in Taiwanese men
Umeizudike *et al.,* (2016) Nigeria Periodontal subjects and its association with self-reported hypertension in non-medical staff in a university teaching hospital in Nigeria	N= 276 subjects Mean age: 41.5 ± 8.9 [23-60] Gender: 147 (53% men) Smoking status: former or non-smoker Systemic health: check up by GP and recorded	CPI (WHO) CPI 3-4: periodontitis	Self-reported	24.4% of patients (33) with HT had periodontitis (CPI codes 3/4) versus 12.1% (17 patients) who had no periodontitis (CPI codes 1/2) p=0.012	Periodontal disease was highly prevalent in this study. Self-reported HT was associated with periodontitis, older age, lower education and a positive family history. Periodic periodontal examination and regular blood pressure assessment for non-medical staff is recommended

Author, year, country, title	Population of study	Periodontal criteria	Blood pressure (BP)	Observed effect (expressed as OR, RR, etc…)	Publication conclusions
Vieira *et al*., (2011) Brazil Severe periodontitis is associated with diastolic blood pressure elevation in individuals with heterozygous familial hypercholesterolemia: a pilot study	N= 79 subjects with heterozygous familial hypercholesterolemia (hFH) Age: [17-80] years Gender: 31 males and 48 females Smoking status: not specified. Systemic health: exclusion criteria: previous cardiovascular events, diabetes mellitus, thyroid diseases, congestive heart failure, pregnancy, renal diseases, any current inflammatory disease, serum calcium disturbances, and use of antibiotics and lipid lowering drugs in the 3 months and 6 weeks, respectively, before the study	-Severe perio: ≥3 sites, not on same tooth, with CAL≥7 mm and ≥1 interproximal site with PPD≥5 mm -Non-severe perio group (moderate periodontitis): ≥3 sites with CAL ≥5 mm, not on the same tooth, and PPD ≥4mm -Gingivitis, and periodontal health (Bassani *et al*. 2007)	BP was measured as previously recommended (Frohlich *et al.* 1995)	Severe perio= 41.8% (N = 33) Non-severe perio=58.2% (N= 46) The association between severe periodontitis and DBP was confirmed after adjustment for traditional atherosclerotic risk factors, i.e., TC, age, smoking, HDL-C, BMI, and BP Only the association between severe periodontitis and DBP (OR= 3.1, 95% CI (1.1-8.5); p= 0.03) was confirmed	In individuals with hFH, severe periodontitis was associated with a higher DBP, which suggests that severe periodontitis, itself, may contribute to the increased cardiovascular risk profile in this population
Yamori *et al*., (2011) Tanzania Hypertension, Periodontal Disease, and Potassium Intake in Non-smoking, Non-drinker African Women on No Medication	N= 81 Tanzanian women Age: 46–58 years range Gender: only women Smoking status: non-smokers Systemic health: not on any medication	CPITN	BP was measured using a centrally calibrated automatic BP measurement system used for the CARDIAC Study	Severity of periodontitis was significantly correlated with SBP (r = 0.288, p=0.018) DBP (r = 0.293, p = 0.015) Severely hypertensive group (BP>180/110 mm Hg) had significant difference in the severity of periodontitis (CPITN, 2.82 ± 0.64 from normal to borderline hypertensive BP> 160/100) group (2.29 ± 0.61, p< 0.05)	Potassium intake may be an important factor linking periodontitis and HT in middle-aged non-smoking and non-alcoholic women on no medication, although chronic inflammation such as periodontitis may cause HT through a more direct mechanism

Author, year, country, title	Population of study	Periodontal criteria	Blood pressure (BP)	Observed effect (expressed as OR, RR, etc…)	Publication conclusions
(Megat Mohd Zainoddin, 2013) Malaysia Retrospective record review study performed at School of Dental Sciences, University Sains Malaysia	N= 370 periodontal patients Mean age: 39.25 ±14.2 Gender: 134 (36% male vs 236 (63% female) Smoking H: Only non-smokers included Systemic health: Recorded as per records of patients notes	Severity of CP was classified according to the probing pocket depth (PPD) as Mild CP= PPD=3-4 mm Moderate CP= PPD=5-6 mm Severe CP= PPD >6 mm	In this study, systemic diseases were also further divided into hypertension (HPT), diabetes mellitus (DM), and combination of both conditions	76.9% (40 patients) of patients with chronic periodontitis had HT vs 23.1% (12 patients) of with gingivitis had HT Significant associations between chronic periodontitis and all systemic conditions (p< 0.001), diabetes mellitus (p= 0.012), hypertension (p< 0.001) and combination of diabetes mellitus and hypertension (p= 0.004) as determined by chi- square test. No significant association between perio severity and systemic conditions	The prevalence of systemic conditions in patients with periodontal disease was 30.5% comprising more of HT and diabetes mellitus. This study found that systemic conditions were significantly associated with chronic periodontitis (p< 0.001) that may highlight the importance of the collaboration between medical and dental practitioners

EVIDENCE TABLE FOR CASE-CONTROL STUDIES

Author, year, country, title	Population of study	Periodontal criteria	Blood pressure (BP)	Observed effect (expressed as OR, RR, etc…)	Publication conclusions
Buhlin *et al.* (2003) Sweden Risk factors for cardiovascular disease in patients with periodontitis	-Periodontitis group (N=50)**:** 29 men, 21 women; mean age: 52.7 years [37–68] Smoking status: 20 patients were smokers (average 13.4 ± 5.8 cigarettes/day) -Non-periodontitis group (N=47): 18 men, 36 women; mean age 50.2 years [36–70] Smoking status: 17 patients were smokers (13.4±5.8 cigarettes/day) Systemic health: Patients with baseline high BP were excluded. No other CVD, infectious, or ongoing systemic disease at any point during study.	Full mouth charting. PPD ≥ 4mm considered disease Inclusion criteria for periodontitis: ≥ 7 sites exhibiting at least 6 mm CAL DX give was periodontitis gravis: horizontal loss of supporting tissue by ≥1/3 of the root-length with BOP, furcation and/or IBD with pus	BP measured throughout study Other measures: total cholesterol, HDL, CRP, IL-6, TNF - receptor 1, elastase-A1AT complex	-Periodontitis group (mean ± SD): SBP = 135 ± 25mm Hg DBP = 79 ± 12mm Hg -Non-periodontitis group: SBP = 141 ± 25mm Hg DBP = 83 ± 11mm Hg Statistical difference between groups: not stated.	Serological differences in subjects with periodontitis, some of which involve established risk factors for atherosclerosis, might provide insight into the reported epidemiological association between periodontitis and cardiovascular disease
Engström *et al*. (2007) Sweden Association between high blood pressure and deep periodontal pockets	N= 1,239 patients Age range: 35-65 years. Mean age: known HT and their referents: 54 years old; with previously unknown high BP and their referents: 49 and 48 years old Gender: 51.8% were female Smoking (recorded by interview). Patients were classified as: never used tobacco, tobacco ex-user, current smoker, or moist snuff user. Systemic health (recorded by interview). 195 cases (with high BP) and 195 healthy referents	Full mouth charting. PPD ≥5 mm (excluding the third molars)	BP measured throughout study. DBP> 90 mm Hg and known HT patients	Subjects with periodontal pockets had 76% higher odds of being a case than those who had no pockets (adjusted OR 1.76, 95% CI 1.14–2.72 for age, sex, smoking habits, snuff taking and number of erupted teeth)	There was an association between diastolic BP and prevalent deep periodontal pockets. Whether the relationship is a causal one remains to be explored

Author, year, country, title	Population of study	Periodontal criteria	Blood pressure (BP)	Observed effect (expressed as OR, RR, etc…)	Publication conclusions
Gorski *et al*. (2016) Poland The association between dental status and risk of acute myocardial infarction among poles: case-control study	Cases= 134 patients hospitalized with acute MI under the age of 70 years. Mean age=54.3 ± 8.1years, 29 women, 105 men Controls= 155 patients drawn from general population with no MI history (age <70 years. Mean age= 54.9 ± 10 years. 94 women, 61 men Smoking status: patients classified in 3 groups: current smokers if they declared smoking ≥10 cigarettes a day continually for ≥ five years, past smokers and never smokers. Systemic disease: social and medical history taken. Physical examinations performed by a single cardiologist	(1) The severity of periodontitis classified in accordance with Page & Eke (2007) (2) The extent of periodontitis determined on the basis of the percentage of sites with CAL ≥ 3 mm (Arbes Index)	Arterial HT was defined as SBP ≥140 mm Hg or DBP ≥ 90 mm Hg	-The extent of periodontitis was significantly associated with the risk of acute MI even after adjusting for age, sex, tobacco smoking, hypertension, diabetes, BMI, education and income (OR = 2.4, 95% CI = 1.1-5.2); p = 0.0203) -The severity of periodontitis was associated with MI after adjusting for age and sex (OR = 2.0, 95% CI = 1.2–3.5; p=0.0109), but not after adjusting for the other above-mentioned risk factors -59.5% (90 people) with extensive periodontitis had HT vs 44.5% (61 people) without periodontitis p=0.0105. -63.6 % (75 people) with severe periodontitis had HT versus 44.7% (76 people) with no severe periodontitis P=0.0016	This study proves the positive association between periodontitis and acute MI in Poles. This association seems to be stronger with regard to the extent rather than to the severity of periodontitis
Houcken *et al*. (2016) Netherlands Arterial stiffness in periodontitis patients and controls. A case–control and pilot intervention study	N: 109 patients total Cases (patients with periodontitis) = 57 patients Mean age: 46.6 years old ± 8.4, 33 males Controls (non-periodontitis) = 48 patients Mean age 45.5 years-old ± 6.6, 25 males Smoking: recorded Systemic disease: Healthy individuals. Excluded patients with any given disease or chronic medical condition (including diabetes mellitus, rheumatoid arthritis, bacterial infections, severe hypertension (4160 systolic and/or 4110 diastolic mm Hg) and body mass index (BMI) 435 or if they took any anti-inflammatory drugs or antibiotics or were o18 years of age or pregnant	Tonetti &Caffley, (2005) proximal attachment loss of ≥ 3 mm in ≥ 2 non-adjacent teeth with BOP	Measured with Tensiomed Kft twice and a mean score used	The mean SBP/DBP: For periodontal patients: 119.9±14.7/75.0±11.7 For controls: 123.5±13.6/78.5±9.1	It can be concluded that periodontitis is associated with increased arterial stiffness. This confirms with a new parameter the association of periodontitis with ACVD. Although periodontal treatment did not lower arterial stiffness significantly, a modest reduction of SBP after 6 months was observed

Author, year, country, title	Population of study	Periodontal criteria	Blood pressure (BP)	Observed effect (expressed as OR, RR, etc…)	Publication conclusions
Jaramillo *et al*. (2017) Colombia Association of metabolic syndrome and chronic periodontitis in Colombians	Case-control Population: Cases: 431 periodontitis patients, 259 females (60.1%), median age: 48 (27–84). Controls: 220 healthy-gingivitis control, 157 females (63.6%); median age: 45 (28–69). Smoking status: Current smoking measured (yes/no) Systemic disease: questionnaire and clinically measured	Page & Eke (2007)	BP ≥130/85 mmHg	HT present in: 20% of cases (86 people) with periodontal disease and 10.5% of controls (23 people) p=0.002 Median SBP= 116.5 mm Hg (80-170) in controls versus 120 mm Hg (90-200) in cases p=0.012. Median DBP= 78 mm Hg (50-110) in controls versus 80 mm Hg (50-112) in cases p=0.128	The study confirmed the positive association between MetS and periodontitis, being glucose sensitivity the strongly associated component
Kumar *et al.* (2016) India Association of chronic periodontitis with metabolic syndrome: A cross-sectional study	N= 259 Mean age: 39.06 years in cases vs. 37.9 in controls Gender: male 46.9% cases vs. 58.9% controls Smoking status: current tobacco consumers: 14.6% cases vs. 11.6% controls Systemic health: Assessment of metabolic syndrome criteria Cases:130 periodontal patients Controls: 129 non-periodontal patients	Full mouth charting (4 sites per tooth (mesio-buccal, buccal, distobuccal, and lingual) Diagnosis of moderate/ severe periodontitis: Armitage, (1999) and Flemming, (1999) Moderate chronic periodontitis: At least 20 teeth and more than 30% of sites with CAL= 3–4 mm Severe chronic periodontitis: 30% of sites with CAL ≥5 mm	BP ≥130/85 mmHg	15.4% (20) of cases (periodontitis) versus 13.2% (17) of controls (non-periodontitis); p=0.741 Mean SBP/DBP higher in cases (125.77/82.99 mm Hg, compared with control (122.81/81.3 mm Hg) Results were not statistically significant	The results of this study suggest that there is a strong association between chronic periodontitis and MeS The association was independent of the various potential confounding risk factors affecting the chronic periodontitis such as age, sex, residential background, and tobacco consumption

Author, year, country, title	Population of study	Periodontal criteria	Blood pressure (BP)	Observed effect (expressed as OR, RR, etc…)	Publication conclusions
Marjanovic & Buhlin (2013) Sweden Periodontal and systemic diseases among Swedish dental school patients – A retrospective register study	Cases: periodontal patients (N= 325), mean age: 61.7 years-old, gender: even group, smoking status: 51.5% were smokers Controls: non-perio (N=149), mean age: 56.2 years-old, gender: 63.8% female rate, smoking status: 63.8%. Smoking status: (EFFICA database) Smokers: currently smoking Non-smokers: never smokers or quit ≥5 years ago Systemic health: amnestic data regarding age, gender, tobacco use, periodontal disease diagnosis and the patient's medical records were obtained from the dental charts	Armitage, (1999)	Self-reported medical information obtained from the dental charts	23.7% (77 patients) with periodontitis had HT vs 12.1% (18 patients) with no periodontitis had HT An association between periodontitis and all CVDs in general. OR= 1.79 (95% CI 1.12–2.86)	This study found that patients with periodontitis presented with more systemic diseases, such as CVD and diabetes mellitus than control patients. However, no association was found between periodontitis and respiratory diseases. At the present time, the reasons for the associations or lack of association are unknown
Turkoglu *et al.* (2014) Turkey Evaluation of systemic levels of neutrophilic enzymes in patients with hypertension and chronic periodontitis	N=95 patients (43 males and 52 females, aged 33 to 75 years; mean age: 50.9 – 9.7 years). Divided in 3 groups: healthy controls (29), HT control (32), HT with CP (34) Age: 1- 43±8 years-old, 2- 52±8 years-old, 3- 55±7 years old Gender: males/females 1- 13/16, 2- 13/19, 3- 17/17 A total of 95 patients Smoking: smoker (≥,5 cigarettes day, non-smoker (never smoked, former smoker (had quit) Systemic health: Exclusion criteria: having systemic disease, such as type 1 or type 2 diabetes, rheumatoid arthritis, autoimmune diseases, and liver disorders	Armitage, (1999)	BP≥140/90 mm Hg or taking antihypertensive medication for at least 2 years	Levels of neutrophilic enzymes MMP-8, MMP-9, MPO, and neutrophil elastase (NE) in circulation were increased in patients with hypertension and CP	The presence of HT along with CP has a considerable effect on serum neutrophilic enzyme levels, except TIMP-1. However, the levels of these enzymes do not seem to be affected by the presence of HT only. Further studies including patients who have only CP might help illuminate the effect of CP on these enzymes in patients with HT

Author, year, country, title	Population of study	Periodontal criteria	Blood pressure (BP)	Observed effect (expressed as OR, RR, etc…)	Publication conclusions
Vidal *et al.* (2011) Brazil Higher prevalence of periodontitis in patients with refractory arterial hypertension: a case–control study	N= 137 Cases: 70 patients (with primary refractory HT) Mean age: 55.2 ± 9.2 years Gender: Male 34.3% Smoking status: 37% were smokers Controls: 67 non-HT subjects Mean age: 50.0 ± 7.2 years Gender: Male 34.3% Smokers: 27% were smokers Systemic disease: All case and control patients were from a low social economical class	Severe periodontitis (Preshaw, 2009): ≥5 sites with CAL≥6 mm Generalized chronic periodontitis (Armitage, 1999): ≥ 30% of the sites with CAL ≥4 mm	Refractory HT BP >140/ 90 mm Hg, even when the patient is engaged in a treatment program and uses three or more classes of anti-hypertensive drugs including a diuretic	-44.3% and 26.9% had generalized chronic periodontitis in the case and control groups, respectively ($p< 0.05$), while 61.4% and 28.4% of the patients were diagnosed as severe chronic periodontitis in the case and control groups ($p< 0.05$) -The results of this study showed that the OR for the association between arterial HT and severe chronic periodontitis and generalized chronic periodontitis were (OR= 4.04, 95% CI (1.92; 8.49) and OR= 2.18, 95% CI (1.04; 4.56), respectively -Gender, race, diabetes, alcohol drinking problems, and smoking were not significant risk indicators for this sample	Severe and generalized chronic periodontitis seem to play a role as risk indicators for HT patients

EVIDENCE TABLE FOR COHORT STUDIES

Author, year, country, title	Population of study	Periodontal criteria	Blood pressure (BP)	Observed effect (expressed as OR, RR, etc…)	Publication conclusions
Hujoel *et al.* (2000) United States Periodontal disease and coronary heart disease risk	N=8,032 from NHANES I Age: 25-74 years Periodontitis patients (N=1,859): 52.4 ± 0.5 years old Gingivitis (N=2,421): 43.0 ± 0.4 years old Healthy periodontium (N=3752): 42 ± 0.5 years old Smoking history: duration in years and packs per day/ packs-years reported Systemic health: baseline information included medical examination, a standardized medical and dental examination, lab test and 24-h dietary recall Cohort: Up to 10 years	Russel Periodontal index 4 levels defined, and dose-response relationships evaluated	Measured in the clinical examinations but no diagnosis given	SBP mean ± SE: -Periodontitis patients: 139.9± 0.7 mmHg -Healthy patients: 127.5 ± 0.6 mmHg p<0.005 DBP mean ± SE: -Periodontology patients: 85.5 ± 0.4 mm Hg -Healthy patients: 80.1 ± 0.3 mmHg p<0.005	This study did not find convincing evidence of a causal association between periodontal disease and CHD
Inoue *et al.* (2005) Japan Association of periodontitis with increased white blood cell count and blood pressure	N= 364 Mean age: 39.8 ± 11.1 years [20 -59 years]. Gender: 271 (74.5%) were male Smoking status: recorded in the medical examination. Systemic health: health screening performed by the Japan Preventive Medicine Association. Medical examination, medical interview, medical history and laboratory tests Cohort: 1 year	CPITN	Measured in the right arm using a normal mercury sphygmomanometer in accordance with the guideline of the health screening	-Four subjects (7.3%) with periodontitis and 16 subjects (5.2%) without periodontitis had current history of treated HT -SBP differences between groups (patients with periodontitis vs no-periodontitis): 8 mm Hg (unadjusted), 6.1 mm Hg (adjusted 1), and 6 mm Hg (adjusted 2) at baseline, and 6.2, 4.2 and 3.7 mmHg at follow-up, respectively. -DBP difference between groups (patients with periodontitis vs no periodontitis): 4.9 mm Hg (unadjusted), 3.6 mm Hg (adjusted 1), and 3.4 mm Hg (adjusted 2) at baseline, and 5.1, 3.7 and 3.7 mmHg at follow-up, respectively Results were all statistically significant except adjusted 1 and 2 for SBP	Periodontitis is associated with increased BP and WBC count. This finding may provide one underlying pathway linking periodontitis and CVD.

Author, year, country, title	Population of study	Periodontal criteria	Blood pressure (BP)	Observed effect (expressed as OR, RR, etc…)	Publication conclusions
Kawabata *et al.* (2016) Japan Relationship between pre-hypertension/ hypertension and periodontal disease	N= 2,588 students Age: 18.2 ± 0.7 years [18–27] Gender: 1,278 males, 1,310 females Smoking status recorded (current/past/ never) Systemic health: the participants included in the study had no self-reported history of HT and no medication history Cohort: 3 years	-CPI -Percentage of BOP (Furuta *et al* 2011) -Levels of dental plaque and calculus were assessed using the simplified oral hygiene index (Greene *et al* 1964)	-HT was defined by SBP≥140 mm Hg or DBP≥90 mm Hg -Pre-hypertension was defined by SBP= 120–139 mm Hg or DBP= 80–89 mm Hg	-Normal BP patients: the risk of having pre-HT/ HT after 3 years was not significantly associated with PPD≥ 4 mm (OR=0.95 (0.54–1.56); p=0.83) or with ≥ 4 mm and BOP ≥ 30% at baseline (OR: 0.93 [0.51–1.70]; p=0.82) -Participants with pre-HT at baseline, the risk of HT was significantly associated with periodontal disease defined as the presence of both probing pocket depth (PPD) ≥ 4 mm and BOP ≥ 30% at baseline (OR: 2.74 (1.19–6.29); p= 0.02) -The risk of pre-HT was not associated with presence of periodontal disease (OR= 0.93 (0.51–1.70); *p* = 0.82)	In the short-term prospective cohort study, a significant association between presence of periodontal disease and HT was observed in Japanese university students
Lee *et al.* (2015) Korea Association of lifestyle-related comorbidities with periodontitis. A nationwide cohort study in Korea	N= 1,025,340 people (representing 2% of the total population) Age: all ages recorded in the screening Gender: 321,103 (31.3%) were diagnosed with periodontitis, consisting of 158,303 males (49.3%) and 162,800 females (50.7%). Smoking status: recorded (national survey) Systemic health: recorded (national survey) Cohort study (2002-2013). Samples analysed retrospectively Cohort: 11 years	-Periodontitis defined as: acute (K052), chronic (K053), periodontosis (K054), other periodontal disease (K055), and unspecified periodontal disease (K056). -Inclusion criteria based on criteria of the AAP as well as ICD-10 classification criteria	Diseases were diagnosed using the Korean Classification of Disease, sixth edition (KCD-6)	The lifestyle related comorbidities (LCs) with periodontitis' group comprised 253,538 patients and had an overall prevalence rate of HT (43.9%), -Association of Lifestyle-Related Comorbidities: HT with Periodontitis: Univariate analysis: OR=1.96 95% CI (1.94 – 1.98) Multivariate analysis: OR=1.07 95% CI (1.05 – 1.08)	Periodontitis is significantly and positively correlated with LCs (except for myocardial infarction) after adjusting for confounding bias. In particular, lifestyle-related diseases, erectile dysfunction, and osteoporosis seem to be intimately related to periodontitis

Author, year, country, title	Population of study	Periodontal criteria	Blood pressure (BP)	Observed effect (expressed as OR, RR, etc…)	Publication conclusions
Lee *et al.* (2017) Korea Association between periodontal disease and non-communicable diseases A 12-year longitudinal health-examinee cohort study in South Korea	N=354,850 Periodontal disease (PD): 200,026 patients, healthy oral status 154,824. Age >40 years old (group ages recorded). Gender: 199,886 men (56.3%) and 154,964 women (43.7%) Smoking status (questionnaire): not smoking or had smoked fewer than 100 cigarettes were classified as non-smokers, while the other subjects were classified as smokers. Systemic health: As per NHIS–HEC (a self-reported questionnaire, anthropometric measurements, and blood laboratory measurements) Cohort study= Data collected for 12 years (2002-2013) from National Health Insurance Service (NHIS) and NHIS-Health Examinee Cohort Cohort: 12 years	Page & Eke (2007)by a general dentist or a periodontitis in the biannual NHIS programme.	HT (Korean Classification of Diseases, 6th revision [KCD-6], codes I10 and I15; corresponding to the International Classification of Disease, 10th revision [ICD-10], codes I10 and I15)	-Univariate logistic regression analysis (unadjusted) showed: Perio→ HT (OR=0.95, 95% CI (0.93–0.96), p <0.001) -In the multivariate logistic regression analysis with adjustment for sex, age, household income, insurance status, residence area health status, and smoking status, PD was significantly positively related to HT (OR = 1.04, 95% CI (1.01–1.07), p<0.014)	This study has demonstrated that the presence of PD is associated with a significantly elevated risk of NCDs in the Korean adult population, especially obesity, osteoporosis, and angina pectoris. Additional studies are required to confirm this association and to establish in detail the role of the inflammatory pathway in the pathogenesis of periodontitis as a triggering and mediating mechanism
Morita *et al.* (2010) Japan A cohort study on the association between periodontal disease and the development of metabolic syndrome	N=1,023 industrial workers in whom all components of MetS were within the standard values at baseline in 2002. Mean age: 37.3 years Gender: 727 males, 296 females Smoking status: questionnaire Systemic health: the health habits described by Belloc and Breslow (1972) were surveyed using a self-completed questionnaire. Cohort: 4 years	CPI CPI codes ≤2 (without a periodontal pocket) CPI code ≥3 (periodontal pocket ≥4 mm)	BP measured: automatic haemomanometer while patients were sitting. HT SBP ≥130 mm/Hg DBP ≥ 85 mm/Hg	The indices of obesity, HT, lipid abnormality, and hyperglycaemia were positive after 4 years in 73 (7.1%), 140 (13.7%), 69 (6.7%), and 10 (1.0%) subjects, respectively. The positive conversions of hypertension and lipid abnormality after 4 years were significantly associated with the presence of periodontal pockets. OR of non-hypertensive becoming hypertensive in the patients with periodontal pockets was (OR=1.5, 95%CI 1.0-2.3)	The presence of periodontal pockets was associated with a positive conversion of MetS components, suggesting that preventing periodontal disease may prevent MetS

Author, year, country, title	Population of study	Periodontal criteria	Blood pressure (BP)	Observed effect (expressed as OR, RR, etc…)	Publication conclusions
Morita *et al.* (2016) Japan Association between the duration of periodontitis and increased cardiometabolic risk factors: a 9-year cohort study	N= 572 adult industrial workers Mean age 37.4 years 417 men and 155 women Smoking status checked by a questionnaire Systemic health: BP, BMI, triglycerides, HDL cholesterol, and fasting blood glucose levels were measured Cohort: 9 years	CPI (10 representative teeth in 6 sextants were examined by a dental hygienist under the supervision of dentists) Non-periodontitis: CPI ≤2 Periodontitis: CPI ≥3	BP measured in a sitting position using an automatic haemomanometer. Hypertension: SBP of ≥130mmHg or DBP of ≥85 mmHg	-At baseline, 89 participants (15.6%) had periodontal pockets, and 483 (84.4%) did not have periodontal pockets -HT in patients with cumulative duration of periodontal pockets for ≤5 years (OR=1.5, 95% CI= (0.9–2.4), and OR=2.2, 95% CI (1.1–4.3) in group ≥6 years, compared to the group without periodontal pockets. Differences were statistically significant in the group with a cumulative duration of periodontal pockets for ≥6 years (P < 0.05) -Adjustments were made for age, sex, cigarette smoking, alcohol consumption, and maintenance of a healthy body weight	CP was significantly associated with having cardiometabolic risk factors during the 9-year observation period, suggesting that the risk of cardiometabolic disease might increase in people who have untreated periodontitis
Rivas-Tumanyan *et al.* (2012) United States Periodontal disease and incidence of hypertension in the health professionals follow-up study	N=31,543 participants of the Health Professionals' Follow-Up Study (HPFS) prospective cohort. This sample contributed to 466,514 person-years during 20 years of follow-up (1986-2006). Age: male health professionals (dentists, pharmacists, optometrists, podiatrists, osteopaths and veterinarians) age 40–75 Smoking status: information on smoking was summarized in the comprehensive smoking index (CSI) Systemic health: validity of self-reported diabetes, physical activity, weight, alcohol, diet and supplement intake measures in this cohort have been demonstrated previously Cohort: 20 years	Self-reported periodontal disease	Self-reported HT	After adjusting for risk factors, we did not observe a significant association between periodontal disease diagnosis at baseline and the risk of incident HT (RR = 1.04, 95% CI [0.98–1.1]). Results were similar for the analysis on periodontal disease during follow-up (RR = 1.01, 95% CI [0.96–1.05]). There was no evidence for a dose–response relation between the periodontal bone loss severity level (no, mild, moderate, severe, as reported on the 1996 questionnaire), and HT (p=0.65); however, there was limited power for this analysis. Compared with men reporting no periodontal bone loss, those with severe periodontal bone loss had a RR= 1.02, 95% CI (0.77–1.35) for incident HT	We did not observe an association between periodontal disease measures and incident hypertension in this cohort of middle-aged men.

Author, year, country, title	Population of study	Periodontal criteria	Blood pressure (BP)	Observed effect (expressed as OR, RR, etc…)	Publication conclusions
Touminen *et al.* (2003) Finland Oral health indicators poorly predict coronary heart disease deaths	N=6,527 men and women from a nationally representative sample who participated in the health examination with a dental check Gender: 3,091 men Age: >30 years old (30-69) Smoking status: recorded as current smokers, quitters, or never-smokers. Systemic health: recorded, interview and health examination	Modified Periodontal Treatment Need System (PTNS) (Johansen et al 1973).	HT: BP≥170/100 mm Hg or taking continuous medication for HT	Prevalence of HT in men>6 mm pockets (759):16.3% vs 11.8% and 10.7% for 4-6 mm pockets (1,272) or none (487 subjects) respectively ($p<0.01$). Prevalence of HT in women>6 mm pockets 10.4% vs 10.5% and 12.8% for those with 4-6 mm pockets (1,152) or none (816 subjects) respectively. The Differences not statistically significant. OR=1, 95%CI [0.6-1.6] perio→Cardiovascular heart disease mortality Adjusted for age, other oral health indicators, level of education, hypertension, hypercholesterolemia, smoking, and diabetes	The associations between oral health indicators and CHD are mostly explained by confounding factors, particularly those relating to health behaviour.

EVIDENCE TABLE FOR INTERVENTIONAL STUDIES

Author, year country, title	Study design	Population characteristics	Intervention	Follow-up	Impact of periodontal therapy on BP levels	Impact of periodontal therapy on biomarkers
Seinost *et al*., (2005) Austria Periodontal treatment improves endothelial dysfunction in patients with severe periodontitis.	Interventional study (non-RCT)	N: 60 individuals Groups: -30 individuals with severe PD (defined as 6 teeth with pocket depth N5 mm and loss of attachment of z3 mm in 3 aspects of each involved tooth) -31 control (no periodontal disease) MH: Otherwise healthy (HT was an exclusion criteria)	-Test Group: OH instructions, multiple supra-gingival cleaning, NST completed in two session, 7 days of amoxicillin plus clavulanic acid and metronidazole -Control Group; No intervention	3 months	No changes in blood pressure There was no statistically significant change in SBP (P = 0.68) and DBP (P = 0.2) at follow-up. Measurements not shown in the manuscript, requested information to the authors by email but no reply obtained	Patients with periodontitis had significantly higher baseline levels of ESRs and concentrations of C-reactive protein than did controls. CRP concentrations significantly decreased and there was a trend toward a reduction in ESRs
D'Aiuto *et al.*, (2006) United Kingdom Periodontal infections cause changes in traditional and novel cardiovascular risk factors: results from a randomized controlled clinical trial	RCT	N:40 individuals with chronic generalized PD (defined as 50% of dentition with PPD>4 mm and with documented radiographic alveolar bone loss) Groups: -Standard periodontal therapy (SPT)= 20 individuals -Intensive therapy (IPT)= 20 individuals MH: Otherwise healthy	SPT: One single session of NST IPT: One single session of NST and LDA (minocycline microspheres)	1, 2, 6 months	Reduction in SBP in the IPT group after 2 months but back to baseline parameters at 6 months: (135±14mmHg→129±17mmHg) P=0.04 A mean difference of 7±3 mm Hg in SBP (95% CI 1-12, P = .0211) for IPT with respect to SPT was observed at 2 months, which was greater for smokers: (14±5 mm Hg 95% CI 3-25, P = 0.0124) No effect on DBP at 2 or 6 months: 86±11→85±12→86±13 P=0.888	IPT patients significant decreased in inflammatory markers (CRP, IL-6, WBC) at 1 and 2 months, and in lipid markers scores (TC and HDL-C) at 2 to 6 months after treatment compared with SPT

Author, year country, title	Study design	Population characteristics	Intervention	Follow-up	Impact of periodontal Therapy	Biomarkers
Tonetti *et al.,* (2007) United Kingdom Treatment of periodontitis and endothelial function	RCT	N: 120 individuals with generalized severe PD ((probing pocket depths of >6 mm and marginal alveolar bone loss of >30%) with 50% or more of their teeth affected). Groups: -59 test -61 control MH: Otherwise healthy	-Test group OHI, single session of NST+ LDA (microspheres of minocycline) and extraction of hopeless teeth -Control group OHI, single session of supra-gingival scaling and polishing	Day 1, 7, 30, 60,180	Mean difference in SBP/DBP in the test group versus control (Day 1). SBP= 4.60 mm Hg (1.07 to 8.13) *p*=0.01 DBP= 2.95 mm Hg (0.42 to 5.49) *p*=0.02 Both values increased, No effect after 6 months.	Levels of CRP protein, IL-6, and the endothelial-activation markers soluble E-selectin and von Willebrand factor and neutrophils were significantly higher in the test than control groups at day 1 CRP levels and neutrophil counts were decreased 6 months after the administration of the therapy in both treatment groups
Higashi *et al.,* (2008) Japan Periodontal infection is associated with endothelial dysfunction in healthy subjects and hypertensive patients	RCT	Protocol 1: N: 52 individuals Groups: -32 individuals with PD (ascertained by self-reported questionnaire) randomly allocated to periodontal therapy (16) or no intervention (16) -20 healthy controls (periodontally and systemically) MH: Otherwise healthy Protocol 2: N: 64 individuals Groups: -26 individuals with PD randomly allocated to periodontal therapy (17) or no intervention (9) -38 periodontally-healthy individuals MH: CAD patients with HT	Protocol 1+ 2: -Periodontal therapy group: OHI, NST and 4-7 days of systemic antibiotic and mouthwash for 24 weeks - Control Group, and no intervention group did not receive periodontal therapy	Protocol 1: 24 weeks Protocol 2: 24 weeks	Changes in BP in treated patients were not statistically significant: Protocol 1: Before ttx: SBP:115.1±10.9 DBP: 66.1± 7.4 After ttx: SBP: 114.6±10.4 DBP: 67.4±7.9 Protocol 2: Before ttx: SBP: 140.1 ± 20.3 DBP: 89.2±14.1 After ttx: SBP: 141.3±21.4 DBP: 88.7±13.7	Protocol 1&2: -Serum concentrations of IL-6 and hs-CRP, indices of systemic inflammation, were significantly higher in patients with periodontitis than in healthy subjects. - The 24 weeks of treatment significantly decreased serum concentrations of IL-6 and hs-CRP. Periodontitis therapy did not alter other parameters (i.e. BP measurement) - Periodontal therapy improves endothelial function through an increase in NO bioavailability

Author, year country, title	Study design	Population characteristics	Intervention	Follow-up	Impact of periodontal Therapy	Biomarkers
Higashi *et al.* (2009) Japan Oral infection-inflammatory pathway, periodontitis, is a risk factor for endothelial dysfunction in patients with coronary artery disease	RCT	N: 101 patients with coronary artery disease randomly allocated to treatment or control Groups: - 48 individuals with chronic PD (self-reported questionnaire and confirmed with oral examination: Defined as the presence of at least 2 teeth with probing pocket depth ≥4 mm and with attachment loss ≥3 mm) - 53 controls (no periodontitis) MH: Coronary artery disease	-Periodontal individuals randomly allocated to periodontal therapy that was NST and 4 to 7 days of antibiotic and mouthwash for 24 weeks (24) or no intervention (24) - Control Group No periodontal treatment Both groups: Intra-arterial infusion of acetylcholine (ACh) and to sodium nitroprusside (SNP) before periodontal therapy in 48 patients with periodontitis and 53 control patients, and in 24 patients who were treated periodontitis and 24 untreated patients before and after 24 weeks of follow- up	24 weeks	Changes in blood pressure were not statistically significant: Test: Before ttx: SBP: 141.3±20.2 DBP: 82.5±13.3 After ttx: SBP: 140.3 ± 19.3 DBP: 80.7 ± 12.9 Control: Before ttx: SBP: 141.3 ± 19.8 DBP: 82.3 ± 12.7 After ttx: SBP: 140.8 ± 19.1 DBP: 81.9 ± 12.1	-Serum concentrations of IL-6 and hs-CRP, were significantly higher in patients with periodontitis than in patients without periodontitis -In patients with periodontitis who received treatment, IL-6 and hs-CRP. Periodontal therapy did not alter other parameters -In the untreated periodontitis group, the baseline clinical characteristics were similar at 0 and 24 weeks of follow-up
Graziani *et al.*, (2010) Italy Systemic inflammation following non-surgical and surgical periodontal therapy	Interventional study (Non-RCT)	N: 14 individuals with generalised severe chronic PD (defined according to Armitage, 1999: PPD ≥5 mm on at least 30% of their dentition) Groups: Only one group No control group MH: Otherwise healthy	OHI, SRP in 2 visits within 24 hours, Widman flap procedure 180 days after completion of NST	Day 1, 7, 30,90,180, 181, 187, 200, 201, 207, 270	The authors reported non-specific changes in arterial BP were observed after both non-surgical and surgical periodontal therapy (as per the Fig. 3 a and b included in their manuscript)	CRP and Serum Amiloid A underwent marked increased 24 h following NST and periodontal surgeries D-dimer levels increased drastically 24 h after NST

Author, year country, title	Study design	Population characteristics	Intervention	Follow-up	Impact of periodontal Therapy	Biomarkers
Taylor *et al.,* (2010) Australia The effect of initial treatment of periodontitis on systemic markers of inflammation and cardiovascular risk: a randomized controlled trial	RCT	N: 125 individuals with PD (defined as ≥6 sites with PPD ≥5 mm and CAL ≥2 other than the third molars) Groups: Randomly allocated to -test (61) -control group (64) MH: Otherwise healthy	-Test OHI, Extractions of hopeless teeth, NST - Control Dental extractions to alleviate pain. No periodontal treatment	3 months	No changes in BP reported in the manuscript. However, authors provided further information: Test: Before treatment: SBP= 135.98± 24,09. DBP= 84,32 ± 12,74 (mean ±SD) After treatment: SBP= 135,08 ± 19,19. DBP= 84,86 ± 10,47 (mean ± SD) Control: Before treatment: SBP= 139.47± 22,99. DBP= 85,52 ± 10,55 (mean ±SD) After treatment: SBP= 134,21 ± 20,02. DBP= 83,70 ± 11,39 (mean ± SD)	-Fibrinogen showed a statistically significant decrease from pretreatment to post-treatment levels within the intervention group - The mean levels of CRP, PAI-1, and vWF showed a tendency to be lower in the intervention group, but this did not reach significance -Hemoglobin, hematocrit and mean cholesterol level showed a statistically significant increase in the intervention group in comparison to the control group over the duration of the study
Lopez *et al*., (2012) Chile Effects of periodontal therapy on systemic markers of inflammation in patients with metabolic syndrome: a controlled clinical trial	RCT	N: 165 individuals with PD (defined as the presence of ≥ 4 teeth with ≥1 site with PPD ≥4 mm and concomitant CAL ≥3 mm) Groups: -Experimental treatment group (ETG)= 82 -Control group (CTG)= 83 MH: MetS	-ETG received plaque control and NST plus amoxicillin and metronidazole -CTG received plaque control instructions, supragingival scaling, and two placebos.	12 months	Authors reported no statistically significant difference between treatment groups in the levels of BP but no mean values were reported in the manuscript Requested authors to send mean values but not reply	-CRP levels decreased with time and that this reduction was significant at 9 (P = 0.024) and 12 (P = 0.001) months in both groups, without difference between the groups -Fibrinogen levels significantly decreased in the ETG at 6 and 12 months but not in the CTG.

Author, year country, title	Study design	Population characteristics	Intervention	Follow-up	Impact of periodontal Therapy	Biomarkers
Vidal *et al*., (2013) Brazil Non-surgical periodontal treatment reduces cardiovascular risk in refractory hypertensive patients: a pilot study	Interventional study (Non-RCT)	N: 26 patients diagnosed with generalised chronic PD (defined according to Armitage, 1999: PPDs) ≥5 mm on at least 30% of their dentition) Groups: Only one group No control groups MH: Refractory hypertension * Blood pressure was assessed using an ambulatory blood pressure monitoring device (TM 2430®, A&D, Santa Clara, CA, USA).	-Oral hygiene instructions -NST at sites with PPD ≥ 4 mm, no time limit	6 months	A significant reduction in the median values of SBP (of 12.5 mmHg) and DBP (of 10 mm Hg) Baseline: SBP: 175 (38.8) DBP: 105 (21.3) After 6-months: SBP: 157.5 (40) DBP: 95 (11.3)	-A significant reduction in the mean values of LVM and PWV was observed. -The median values of the inflammatory markers, CRP, IL-6 and fibrinogen were significantly reduced 6 months after periodontal treatment.
Albush *et al*. (2013) Syria Effect of surgical and non-surgical periodontal debridement on vascular thrombotic markers in hypertensives	Interventional prospective pilot study (Non-RCT)	N: 40 individuals with moderate to severe periodontitis defined as ≥4 teeth with ≥1 site with PPD ≥4 mm and/or CAL ≥3 mm. Groups: -NST group: 20 individuals (PPD=4-6 mm): -Surgical group: 20 individuals (PPD>6mm): MH: HT under control with different antihypertensive medication, non- smokers	NST group (PPD=4-6 mm): Received 1 session of full mouth debridement with ultrasonic and curettes under LA. -Surgical group (PPD>6mm): 1 session of supragingival debridement and Modified Widman Flap. -Blood samples taken at baseline (16 h fasting prior to test), 48 h and 6 weeks post-treatment.	6 weeks	Blood pressure measured at baseline but not after treatment Biomarkers: Platelets, Fib levels, DD levels and vWF:Ag measured.	-No statistically significant differences in vascular thrombotic markers between the two groups at baseline, ($P > 0.05$). -Platelets count, Fib levels and DD levels increased after 48 h of treatment in both groups and decreased after 6 weeks ($P < 0.05$), with no significant differences between the two groups at both time intervals, ($P > 0.05$) -vWF:Ag activity increased significantly in both groups after 48 h ($P < 0.05$) and it decreased after 6 weeks retaining higher values in comparison to baseline with no significant differences between groups at both time intervals, ($P > 0.05$).

Author, year country, title	Study design	Population characteristics	Intervention	Follow-up	Impact of periodontal Therapy	Biomarkers
Houcken *et al*. (2016) Amsterdam Arterial stiffness in periodontitis patients and controls A case-control and pilot intervention study	Pilot intervention study	N:45 patients with PD out of 109 that participated in the cross-sectional study Groups: Only one group with PD (defined as Tonetti &Caffley, 2005= proximal attachment loss of ⩾ 3 mm in ⩾ 2 non-adjacent teeth with bleeding on probing) MH: Healthy otherwise	Treatment: OHI + NST in 2 sessions Twenty patients (randomly assigned) received a systemic antibiotic therapy adjunctive to the scaling and root planning (combination of Amoxicillin 375mg t.i.d. and Metronidazole 500mg t.i.d. for 7 days).	6 months	SBP was significantly reduced 6-months after periodontal therapy 119.8±14.6 to 116.9±15.1mmHg (p=0.04) after adjustment for potential cofounders. DBP: 74.9 ± 11.8 at baseline → 73.1 ± 10.6 6-months after therapy (p=0.05) after adjustment for potential cofounders. For the parameters SBP, DBP and heart rate no other results than those presented were obtained if subgroups for antibiotics usage were analysed	Plasma lipid markers (total cholesterol, HDL, LDL and triglycerides) were assessed. From the biochemical analyses it was clear that the periodontal treatment neither affected levels of total cholesterol nor LDL or HDL and triglycerides
Zhou *et al*., (2017) China Effect of intensive periodontal therapy on blood pressure and endothelial microparticles in patients with prehypertension and periodontitis: a randomized controlled	RCT	N: 95 patients with PD (Page & Eke, 2007) Groups: Randomly assigned to test and control groups -Control treatment group (CTG): 47 -Intensive treatment group (ITG): 48 MH: Pre-hypertension (SBP/DBP= 120–139/80–89 mm Hg)	-Intensive treatment group (ITG): OHI, NST+ locally delivered minocycline hydrochloride ointment (once a week for 4 weeks). Additionally, teeth that could not be saved were extracted. -Control - treatment group only supragingival ultrasonic scaling and polishing at baseline.	6 months	Mean BP was markedly reduced in the ITG compared to the CTG = (absolute difference SBP/DBP: 12.57 mm Hg/9.65 mm Hg, 95% CI: 10.45 to14.69 and 7.06 to 12.24; P <0.05).	-Periodontal treatment lowered the levels of EMPs. -Endotelial microparticles (EMPs) = absolute difference between ITG and CTG= 581.59/µl; 95% CI: 348.12 to 815.06 /µl; P <0.05 -hs-CRP and IL-6 levels were significantly lower in the ITG than the CTG 6 months after intensive periodontal

Author, year country, title	Study design	Population characteristics	Intervention	Follow-up	Impact of periodontal Therapy	Biomarkers
D'Aiuto *et al*., (2018) The UK Systemic effects of periodontitis treatment in patients with type 2 diabetes: a 12 month, single-centre, investigator-masked, randomised trial	RCT	N: 264 individuals with moderate-severe PD (≥20 periodontal pockets with probing pocket depths of >4 mm and marginal alveolar bone loss of >30%), and at least 15 teeth Groups: -133 test (IPT) -131 control (CPT) MH: Type 2 diabetes (WHO) Exclusion criteria were uncontrolled systemic diseases other than diabetes (cardio vascular diseases [including hypertension]	IPT= Initial single session of whole mouth scaling of the root surfaces. 2 months after patients with (plaque scores of ≤20%) and at least one PPD⩾6 mm had periodontal surgical therapy followed by repeated scaling every 3 months CPT= cleaning and polishing the part of the tooth that is visible above the gingiva of all dentition at the same time-points as the IPT group (after baseline and at 2, 6, 9, and 12 months after the completion of the first session of periodontal therapy)	12 months	Mean changes in blood pressure between the two groups were not statistically significant. **IPT:** mean SBP(SD)/DBP(SD): Baseline: 135(16)/ 82(10) Mean SBP[SE]/DBP[SE]: 2M:131[1.4]/80[0.9] 6M: 131[1.4]/81[0.8] 12M: 130 [1.4]/79[0.9] **CPT** (mean SBP/DBP): Baseline: 136(17)/ 83 (9) Mean SBP[SE]/DBP[SE]: 2M: 132[1.4]/80[0.9] 6M: 133[1.4]/80[0.8] 12M: 133[1.4]/81[0.9]	HBA1C, CRP were significantly lower in the IPT group than in the CPT group at 2 months, 6 months, and 12 months TNFα was significantly lower in the IPT group than in the CPT group at 2 months and 12 months Soluble E-selectin and P-selectin were significantly lower in the IPT group than in the CPT group at 6 months Patients in the IPT group had greater FMD at 6 months and 12 months than patients in the CPT group

Appendix 4. Bias Assessment

RISK OF BIAS ASSESSMENT FOR ALL THE STUDIES DESIGNS

Study (first author)	Study design	Selection				Comparability	Outcome/Exposure		Total number of stars (maximum 9)
		Representativeness of the sample	Sample size	Non-respondents	Ascertainment of exposure	Based on design and analysis	Assessment of outcome	Statistical test	
Ahn et al. 2015	Cross-sectional	*	*	*	*	**	*	*	7
Al-Emadi et al. 2006	Cross-sectional			*	*	**	*	*	6
Angeli et al. 2003	Cross-sectional			*	*	*		*	4
Aoyama et al. 2017	Cross-sectional		*	*	*	**		*	6
Arowojolu et al. 2016	cross-sectional			*	*	*	*	*	5
Benguigui et al. 2010	Cross-sectional	*	*	*	*	**	*	*	8
Beukers et al. 2017	cross-sectional	*	*	*		**	*	*	7
Buhlin et al. 2002	Cross-sectional	*	*			**	*	*	6
Carvahlo-Goulart et al. 2017	cross-sectional				*	*		*	3
Chauhan et al. 2016	Cross-sectional	*		*	*			*	4
Chen et al. 2016	Cross-sectional	*			*	**		*	5
Choi et al. 2015	Cross-sectional	*	*	*	*		**	*	7
Chrysanthakopoulos & Chrysanthakopoulos (2016)	Cross-sectional	*		*	*	**	*	*	7
D'Aiuto et al. 2008	Cross-sectional	*	*	*	*	**	**	*	9

Study (first author)	Study design	SELECTION				Comparability	Outcome/Exposure		Total number of stars (maximum 9)
		Representativeness of the sample	Sample size	Non-respondents	Ascertainment of exposure	Based on design and analysis	Assessment of outcome	Statistical test	
Desvarieux et al. 2010	Cross-sectional	*	*	*	*	**	**	*	9
Franek et al. 2009	Cross-sectional	*		*	*	**	**	*	8
Franek et al. 2010	Cross-sectional	*		*	*	**	**	*	8
Fukui et al, 2012	Cross-sectional	*		*	*	**		*	6
Furuichi et al. 2003	Cross-sectional	*		*	*	**	**	*	8
Gomes-Fihlo et al. 2016	Cross-sectional	*	*	*	*	**	**	*	9
Gorska et al. 2017	Cross-sectional	*		*	*			*	4
Han et al. 2010	Cross-sectional	*	*	*	*	**	*	*	7
Han et al. 2016	Cross-sectional	*	*	*		**	*	*	7
Holmlund et al., 2006	Cross-sectional	*		*	*	**	*	*	6
Iwashima et al, 2014	Cross-sectional	*	*	*	*	**	**	*	9
Khocht, 2017	Cross-sectional	*			*	**	**	*	7
Kushiyama et al. 2009	Cross-sectional	*		*	*	**	**	*	8
Kwon et al. 2011	Cross-sectional	*	*	*	*	**	**	*	9
Lysek et al. 2016	Cross-sectional			*	*	**	**	*	7
Machida et al. 2014	Cross-sectional	*		*		**		*	5

Study (first author)	Study design	SELECTION				Comparability	Outcome/Exposure		Total number of stars (maximum 9)
		Representativeness of the sample	Sample size	Non-respondents	Ascertainment of exposure	Based on design and analysis	Assessment of outcome	Statistical test	
Moghadam et al. 2016	Cross-sectional	*	*	*		**		*	6
Morita et al. 2009	Cross-sectional	*		*	*	**	**	*	8
Nesbitt et al. 2010	Cross-sectional			*	*	**	*	*	6
Nesse et al., 2010	Cross-sectional	*		*	*		*		4
Ollikainen et al., 2014	Cross-sectional	*	*	*	*	*	**	*	8
Ribeiro et al. 2016	Cross-sectional	*	*	*	*	**	*	*	8
Rivas-Tumanyan et al., 2013	Cross-sectional	*		*	*	**	**	*	8
Shamsuddin et al., 2015	Cross-sectional	*	*	*	*		*		5
Shimazaki et al. 2007	Cross-sectional	*		*	*	**	*	**	8
Thanakun et al .2014	Cross-sectional	*		*	*	**	**	*	8
Tsakos et al. 2010	Cross-sectional	*	*	*	*	*	**	*	8
Tu et al, 2013	Cross-sectional	*	*	*	*	**	**	*	9
Umeizudike et al. 2016	Cross-sectional			*	*	**	*	**	7
Vieira et al., 2011	Cross-sectional			*	*	**	**	*	7
Yamori et al. 2011	Cross-sectional			*	*		**	*	5
Zainoddin et al, 2013	Cross-sectional			*	*		*		3

Study (first author)	Study design	SELECTION				Comparability	Outcome/Exposure		Total number of stars (maximum 9)
		Representativeness of the sample	Sample size	Non-respondents	Ascertainment of exposure	Based on design and analysis	Assessment of outcome	Statistical test	
Gordon et al. 2018	Cross-sectional	*	*	*	*	**	*	*	8
Alade et al. 2018	Cross-sectional			*	*	**		*	5
Han et al. 2018	Cross-sectional	*	*	*	*	**	*	*	8
Koo et al. 2018	Cross-sectional	*	*	*	*	**	*	*	8
Pietropaoli et al. 2018	Cross-sectional	*	*		*	**	**	*	8

Study (first author)	Study design	Case definition adequate?	Representativeness of the cases?	Selection of controls?	Definition of controls?	Based on design and analysis?	Ascertainment of exposure	Same method for ascertainment for cases and controls	Non-response rate	Total Number of stars. (maximum 9)
Buhlin et al. 2003	Case-control	*			*	**	*	*	*	7
Engström et al. 2007	Case-control	*	*	*	*	**	*	*	*	8
Gorski et al. 2016	Case-control	*	*	*	*	**	*	*	*	9
Houcken et al. 2016	Case-control	*	*	*	*	**	*			7
Jaramillo et al 2017	Case-control	*		*	*		*	*	*	6
Kumar et al. 2016	Case-control	*		*			*			3
Marjanovic et al., 2013	Case-control	*		*	*	**	*		*	7
Turkoglu, 2014	Case-control	*	*		*	*	*	*	*	7
Vidal et al. 2011	Case-control	*			*	**	*		*	6

Study (first author)	Study design	Representativeness of the exposed cohort	Selection of the non-exposed cohort	Ascertainment of exposure	Demonstration outcome of interest not present at start of study	Based on the design or analysis	Assessment of outcome	Was follow-up long enough for outcome to occur	Adequacy of follow-up cohorts	Total Number of stars. Maximum 9
Hujoel et al. 2000	Cohort	*	*	*	*	**	*	**		8
Inoue et al. 2005	Cohort	*	*	*		**	*			6
Kawabata et al., 2016	Cohort		*	*	*	**	*		*	7
Lee et al., 2015	Cohort	*	*	*	*	**	*	*	*	9
Lee et al. 2017	Cohort	*	*	*	*	**	*	*	*	9
Morita et al, 2010	Cohort		*	*	*	**	*		*	7
Morita et al. 2016	Cohort		*	*	*	**	*	*		7
Rivas-Tumanyan et al., 2012	Cohort		*			**		*		4
Touminen et al. 2003	Cohort	*	*	*	*	**	*	*		8

Study (first author)	Study design	Bias arising from the randomization process	Bias arising from the intended interventions	Bias due to missing outcome data	Bias in measurement of the outcome	Bias in the selection of the reported result	Overall risk of bias
D'Aiuto et al., 2006	RCT	Low	Low	Low	Low	Low	Low
Higashi et al., 2008	RCT	High	Some concerns	Low	High	Some concerns	High
Higashi et al., 2009	RCT	Some concerns	Some concerns	High	Some concerns	Low	High
Lopez et al. 2012	RCT	Low	Low	Low	Low	Low	Low
Taylor et al. 2010	RCT	Low	Some concerns	Low	Low	Low	Some concerns
Tonetti et al. 2007	RCT	Low	Low	Low	Low	Low	Low
Zhou et al. 2017	RCT	Low	Low	Low	Low	Low	Low
D'Aiuto et al. 2018	RCT	Low	Low	Low	Low	Low	Low

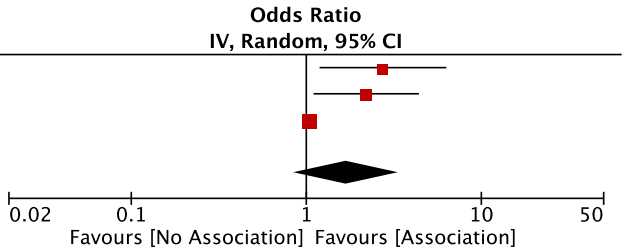
Study (first author)	Study design	Pre-intervention		At intervention	Post-intervention				Overall risk of bias
		Confounding	Selection of participants	Classification of interventions	Deviation from intended interventions	Missing data	Measurement of outcome	Selection of the reported result	
Albush et al. 2013	Interventional Non-RCT	Serious	Moderate	Moderate	Low	Moderate	Moderate	Low	Serious
Graziani et al. 2010	Interventional pilot one group only study	Study design not subjected to bias assessment							
Seinost et al., 2005	Interventional	Moderate	Low	Low	Low	Moderate	Low	Low	Moderate
Vidal, 2013	Interventional prospective cohort study	Study design not subjected to bias assessment							
Houcken et al. 2016	Cross sectional pilot interventional one group study	Study design not subjected to bias assessment							

Appendix 5. Forest Plot Cohort Studies

Study or Subgroup	log[Odds Ratio]	SE	Weight	Odds Ratio IV, Random, 95% CI	Year
Kawabata et al. 2016	1.008	0.4255	26.5%	2.74 [1.19, 6.31]	2016
Morita et al. 2016	0.7885	0.3537	30.1%	2.20 [1.10, 4.40]	2016
Lee et al. 2017	0.0392	0.0149	43.4%	1.04 [1.01, 1.07]	2017
Total (95% CI)			**100.0%**	**1.68 [0.85, 3.35]**	

Heterogeneity: $Tau^2 = 0.28$; $Chi^2 = 9.64$, df = 2 (P = 0.008); $I^2 = 79\%$

Test for overall effect: Z = 1.49 (P = 0.14)



Appendix 6. Prevalence of Hypertension

AUTHOR/YEAR	PREVALENCE OF HYPERTENSION IN PATIENTS WITH AND WITHOUT PERIODONTITIS
Al-Emadi et al. 2006	About 34% of subjects (n=72) with moderate to severe alveolar bone loss reported a history of hypertension, whereas fewer than 8% of patients (n=16) with no or mild alveolar bone loss reported having the disease
Desvarieux et al., 2010	The prevalence of hypertension among participants defined as "healthy" or having either moderate or severe periodontitis was 72%, 58% and 66% (p for linear trend=0.64), respectively
Inoue et al., 2005	Four subjects (7.3%) with periodontitis and 16 (5.2%) subjects without periodontitis had current history of treated hypertension
Lysek et al. 2016	58.3% of people with CPI=3+4 had peripheral BP ≥140/90 mmHg versus 25.5% of people with CPI=1+2 p<0.001 also 62% of people with CPI=3+4 had central BP ≥130/90 mmHg versus 31.4% of people with CPI=1+2 p=0.002
Machida et al, 2014	84 of moderate-severe periodontitis patients (28.7%) had HT vs 17 of non-periodontitis or mild periodontitis (14.0%) had HT p=0.002
Marjanovic et al., 2013	23.7% (77 patients) with periodontitis had HT vs 12.1% (18 patients) with no periodontitis had HT
Nesse et al., 2010	In the dental clinic and the periodontitis clinic the prevalence of HT was significantly higher in patients with periodontitis 13.4% (29 patients) compared to controls 5% (16 patients) (P = 0.001)
Rivas-Tumanyan et al., 2013	64% of the patients with severe periodontal disease had HT versus 70% of patients without severe periodontitis. Additionally, 75% of patients with severe periodontitis presented with high blood pressure vs 55% of those without severe periodontitis
Shamsuddin et al., 2015	12.2% of patients with periodontitis had HT
Megat Mohd Zainoddin, 2013	76.9% (40 patients) of patients with chronic periodontitis had HT vs 23.1% (12 patients) of with gingivitis had HT
Benguigui et al. 2010	The prevalence of HT among participants defined as no periodontitis, moderate or severe periodontitis was 14 (25.9%), 38 (37.2%) and 48 (48.5%) respectively p= 0.021
D'Aiuto et al., 2008	Among individuals with moderate and severe periodontitis, there was a higher prevalence of HT (51–56%, 95% CI 47– 64) vs individuals with no or mild periodontitis (27%, 95% CI 25–29)
Fukui et al, 2012	The percentage of patients with HT was 34.6% and 28.0% for moderate/severe vs non/mild periodontitis respectively (p=<0.001). According to CAL, was 32.1% vs 27.9% for moderate/severe vs non/mild periodontitis respectively (p=<0.001)
Gomes-Fihlo et al. 2016	62.80% (91 patients) with periodontitis had HT versus 67.60% (184 patients) without periodontitis had HT p=0.32 Nevertheless, when BP was measured, 70.30% (102) patients with periodontitis had SBP≥130 vs 59.10% (162 patients) with non-periodontitis p=0.02 and 49.70% (72 patients) with periodontitis had DBP≥85 vs 43.10% (118 patients) without periodontitis
Gordon et al. 2018	16.2% of patients with severe periodontitis had HT versus 26.2% with none or mild periodontitis had HT
Han et al. 2010	48.7% (131 patients) with CPI 3-4 had HT versus 51.3% (138 patients) with CPI 2-1 and HT
Han et al. 2016	38.9%(0.9SD) of the patients with periodontitis had HT versus 20.8%(0.5SD) of the non-periodontal patients had HT p<0.0001
Jaramillo et al. 2017	20% of cases (86 people with periodontitis) had HT versus 10.5% of controls (23 people with no periodontitis) P=0.002
Koo et al. 2018	43% of patients with periodontitis had HT versus 40% of non-perio group *p* <0.001
Kumar et al. 2016	15.4% (20) of cases (periodontitis) versus 13.2% (17) of controls (non-periodontitis) p=0.741
Kwon et al. 2011	36.2% (33.9–38.6) of patients with periodontitis (1167) had HT vs 18.5% (17.1–20.1) of non-periodontitis (718) had HT
Morita et al. 2009	43.2% (277 subjects) with periodontitis had HT versus 30% (549 subject) without periodontitis had HT
Morita et al. 2010	23.4% (48 patients) of patients with pockets developed HT vs 11.3 % (92 patients) without pockets developed HT
Shimazaki et al. 2007	56.8% (21 patients) with CAL>3mm had HT vs 43% (235 patients) with CAL<3 had HT 53% of patients with PPD ≥2 mm had HT vs 41.9% of patients with PPD<2mm
Beukers et al. 2017	12.3% of patients with periodontitis had HT versus 4.2% of non-periodontitis patients. The difference and CI= 8.1% (7.4% to 8.7%)

Goulart et al. 2017	42.6% of periodontitis patients (29 patients) had HT vs 34.1% (93 patients) of gingivitis patients vs 28.8% (57patients) of patients with no periodontal disease
Gorski et al. 2016	59.5% (90 people) with extensive periodontitis had HT vs 44.5% (61 people) with no extensive periodontitis p=0.0105. Additionally, 63.6 % (75 people) with severe periodontitis had HT versus 44.7% (76 people) with no severe periodontitis P=0.0016
Touminen et al. 2003	Prevalence of HT in men with more than 6 mm (759) pockets was 16.3% vs 11.8% and 10.7% for those with 4-6 mm (1272) pockets or none (487 subjects) respectively. The differences were statistically significant p<0.01
	Prevalence of HT in women with More than 6 mm (424) pockets was 10.4% vs 10.5% and 12.8% for those with 4-6 mm (1152) pockets or none (816 subjects) respectively. The differences were not statistically significant
Ribeiro et al. 2016	51.9% (28 patients) of individuals with destructive periodontal disease had HT vs 48.1% (26 patients) of patients with no destructive periodontal disease
Umeizudike et al. 2016	24.4% (33 patients) of patients with Periodontitis (CPI codes 3/4) had HT versus 12.1% (17 patients) who had no periodontitis (CPI codes 1/2) had HT

Prevalence of hypertension in patients with/without periodontitis

AUTHOR/YEAR	PREVALENCE OF PERIODONTITIS IN PATIENTS WITH AND WITHOUT HYPERTENSION
Ahn *et* al. 2015	38.4% of people with HT (≥90/140) (1984 people) had periodontitis (CPITN 3/4) vs 35.2% (1820 people) without HT had periodontitis
Aoyama *et* al. 2017	PPD of 71–80-year-old men with HT was deeper than that of non-HT subjects. There was no statistical difference of CAL
Choi *et* al, 2015	42.4% (SE=1) of subjects with HT had periodontitis (CPI 3/4) vs a 23.9% (SE=0.7) of non-HT patients had periodontitis
Gorska *et* al. 2017	29.3% ± 25.4 of patients with HT had PPD≥4mm vs 23.4% ± 21.9 who didn't have HT had PPD≥4mm
Iwashima *et* al, 2014	23.5% of people with HT had a CPITN4 versus 16.8% of the people that were not HT, and this figure was statistically significant
Vidal *et* al., 2011	44.3% and 26.9% had generalized chronic periodontitis in the refractory arterial HT and control groups, respectively (P < 0.05), while 61.4% and 28.4% of the patients were diagnosed as severe chronic periodontitis in the refractory arterial HT and control groups (P < 0.05)
Engstrom *et* al., 2007	57.1% of patients with high DBP had periodontal pockets vs referents (No high DBP) that had periodontal pockets 42.6% p<0.05. Additionally, 54.7% of known HT patients had pockets vs 38.9% of referents that didn't this was not statistically significant

Prevalence of periodontitis in patients with/without hypertension

Appendix 7. Systolic and Diastolic Blood Pressure values

Author/ year	Periodontitis			Periodontally healthy			*p* value periodontitis vs. no periodontitis		Explanation
	N	SBP±SD	DBP±SD	N	SBP±SD	DBP±SD	SBP	DBP	
Angeli *et al*., 2003	15	172±16	101±11	7	143±22	94±5	p<0.01	p<0.05	Mean ± SD
Benguigui *et al.* 2010	99	136±19.7	83±10	54	125±16.7	78.4±9.4	0.002	0.023	Mean ± SD
Buhlin et *al.*, 2003	50	135±25	79±12	46	141±25	83±11	N/A	N/A	Mean ± SD
Chauhan *et al.* 2016	25	126±8.41	84±9	34	125±3.9	81.4±2.87	NS	NS	Mean ± SD
Desvarieux et al., 2010	248	141	81	53	141	76	0.26	0.001	Mean only
D'Aiuto *et al.*, 2008	337	130 (126.9–132.3)	76.9 (75.2–78.6)	11758	119 (118.2–119.7)	73.6 (73.2–4.0)	N/A	N/A	Mean (CI)
Franek et al. 2009	50	128±9	82±7	49	131±11	85±10	0.29	0.12	Mean ± SD
Franek *et al.* 2010	22	145±11	84±7	14	130±14	79±8	<0.005	0.03	Mean ± SD
Furuichi *et al.* 2003	N/A	134±21.6	78.8±11	N/A	132±22.8	77.4±12	NS	NS	Mean ± SD
Gorski et al. 2016	119	138±19.1	80.8±10	170	129±18.4	80.6±10.9	0.002	0.727	Mean ± SD
Han et al. 2018	1968	113(0.31)	74(0.22)	6373	110(0.18)	72.3(0.15)	<0.001	<0.001	Mean (SE)
Higashi *et al.*, 2008	32	115±10.2	66.1±7	20	115±9.7	65.6±7	NS	NS	Mean ± SD
Higashi *et al.*, 2008	26	141±18.1	89.8±12	38	139±17.5	88.9±11.9	NS	NS	Mean + SD
Higashi *et al.*, 2009	48	141±17.6	82.4±12	53	140±16.8	82.7±10.9	NS	NS	Mean + SD
Houcken *et al.* 2016	57	120±14.7	75±12	48	124±13.6	78.5±9.1	NS	NS	Mean + SD
Hujoel et al. 2000	1859	140 (0.7)	86.5 (0)	3752	128 (0.6)	80.1±0.3	<0.005	<0.005	Mean (SE)
Inoue *et al.*, 2005	55	127±1.8	74.9±1	309	121±0.8	71.5±0.5	0.003	0.017	Mean ± SD
Inoue *et* al., 2005	55	127±2	79.3±2	309	123±0.8	75.6±0.6	0.81	0.02	Mean ± SD
Jaramillo *et al.* 2017	431	120 [90-200]	80 [50-112]	220	117 [80-170]	78 [50-110]	0.012	0.128	Median [range]
Koo et al. 2018	6598	124±17.3	77.1±10.5	6598	122±16.8	76.8±10.1	<0.001	0.15	Mean ±SD
Kushiyama *et al.* 2009	316	130 [89-188]	76 [40-100]	754	124 [80-187]	72 [39-108]	<0.001	<0.001	Median [range]
Kumar *et al.* 2016	130	126±17.1	83±9	129	123±13.8	81.3±8.36	0.126	0.127	Mean ± SD
Kwon *et al.* 2011	3127	120 (119.11–121.14)	76.9 (76.29–77.58)	3709	111 (110.15–111.53)	73.6 (73.07–74.16)	N/A	N/A	Mean (CI)
Lee *et al.* 2017	200026	126±17.4	79.5±12	154824	128±18.8	79.4±11.8	<0.001	0.334	Mean ± SD
Lysek *et al.* 2016	48	145±21.5	87.4±12	51	129±19.1	78.8±10.3	0.001	0.001	Mean ± SD
Morita *et al.* 2009	641	123±17.2	79.3±12	1837	118±16	75.2±12.1	<0.01	<0.01	Mean ± SD
Nesbitt *et al*, 2010	43	134±22.8	79.8±14	157	124±21.9	78.1±9.6	0.01	0.47	Mean ± SD
Pietropaoli et al. 2018	417	134+21.3	71.1±14.7	1734	131±19.5	69.6±13.9	<0.001	0.031	Mean ± SD
Ribeiro et al. 2016	90	124±20.3	76.9±15	135	118±16.5	75.1±15.9	0.01	0.39	Mean ± SD
Shimazaki *et al.* 2007	37	131±22.8	76.1±10	547	127±20.6	76.4±10.4	NS	NS	Mean ± SD
Thanakun et al. 2014	58	133 (118.8, 140.3)	85.5 (78.0, 91.3)	67	120 (112.0, 130.0)	78 (72, 83)	0.001	<0.001	Median (1st, 3rd quartile)

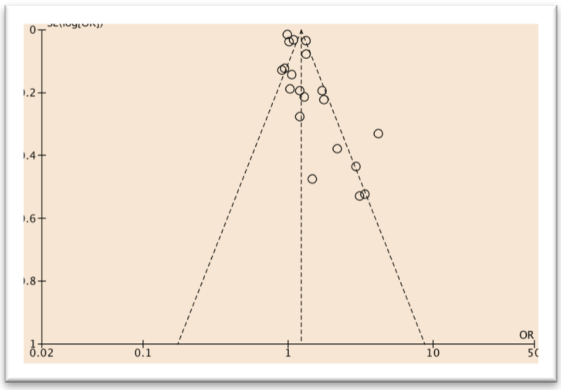
Tu et al, 2013	5650	122±16.3	71.7±10	9870	118±16.3	69.7±10.1	N/A	N/A	Mean ± SD
Tu *et al*, 2013	4377	127±14.9	75.6±10	8638	126±14.6	75.4±9.96	N/A	N/A	Mean ± SD
Vieira et al., 2011	33	121±17.3	80.6±9	46	120±15.1	76.5±8.24	0.88	0.03	Mean ± SD

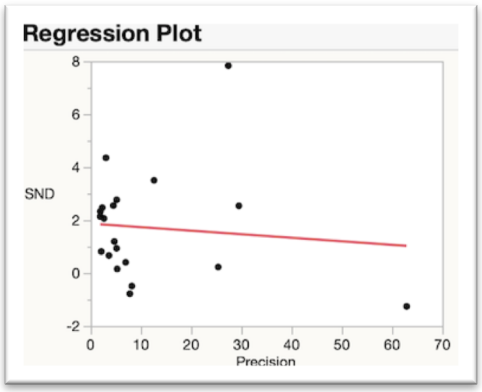
Appendix 8. Publication Bias

Publication bias (Meta-analyses A): OR for hypertension in patients with Periodontitis

Funnel Plot (a) and Egger's test (b) for Cross-sectional and Case-control Studies (Moderate-severe adjusted):

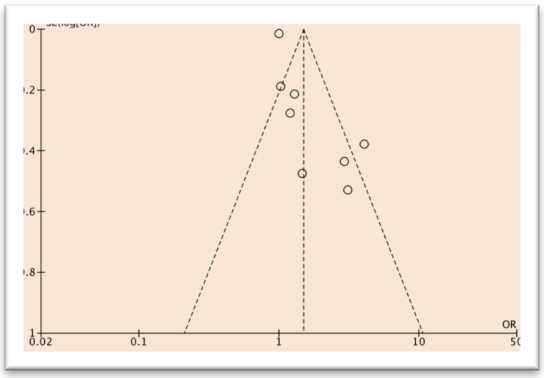
a) **b)**

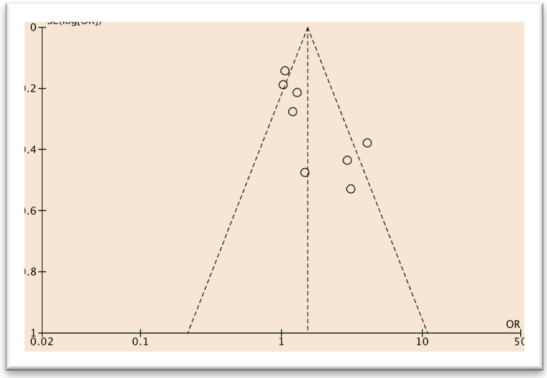




Funnel Plot for Cross-sectional and Case-control Studies (Severe only adjusted) (a) and (Confident case definition adjusted) (b):

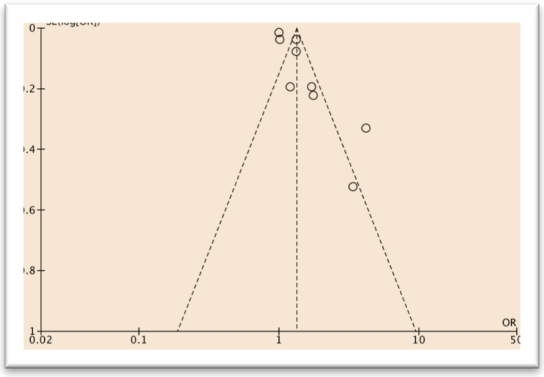
a) **b)**

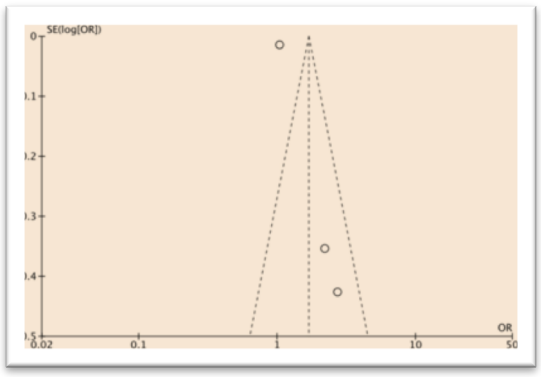




Funnel Plot for Cross-sectional and Case-control Studies (Non-confident case definition adjusted) (a) and (Cohort studies) (b):

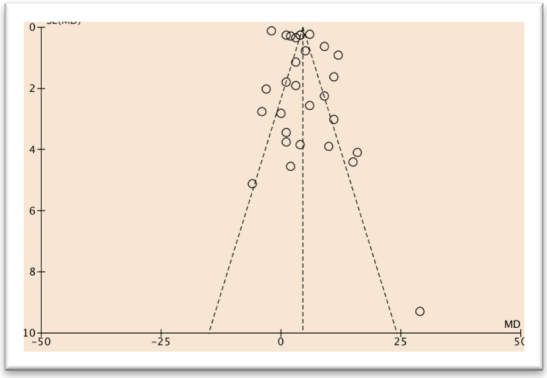
a) **b)**

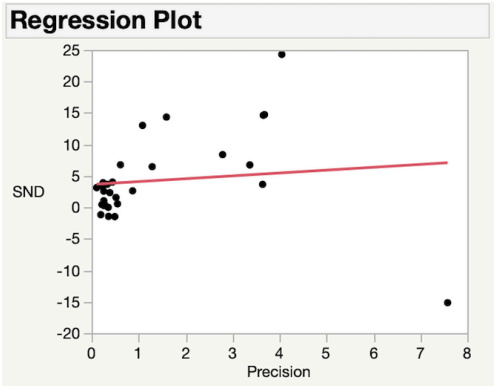




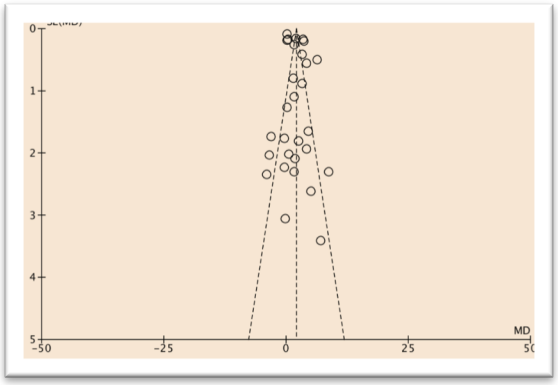
Publication bias (Meta-analyses A): OR for hypertension in patients with Periodontitis

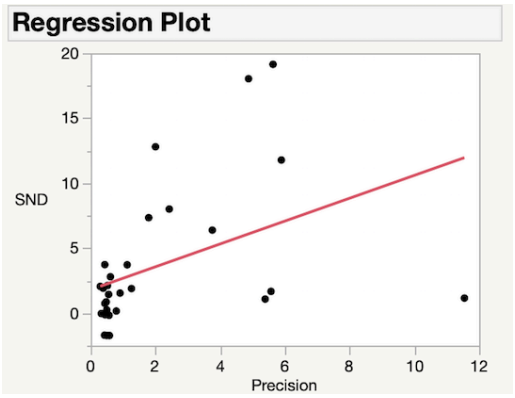
Funnel Plot for all studies showing mean SBP in patients with periodontitis versus patients without periodontitis:





Funnel Plot for all studies showing mean DBP in patients with periodontitis versus patients without periodontitis:





The 2nd original publication

JIM Original Article

doi: 10.1111/joim.13180

Is systemic inflammation a missing link between periodontitis and hypertension? Results from two large population-based surveys

E. Muñoz Aguilera[1,2†], Y. Leira[1,3†], Q. Miró Catalina[4], M. Orlandi[1], M. Czesnikiewicz-Guzik[5,6], T. J. Guzik[7,8], A. D. Hingorani[9], J. Nart[2] & F. D'Aiuto[1]

[1]*Periodontology Unit, UCL Eastman Dental Institute and Hospital, University College London, London, UK;* [2]*Department of Periodontology, Universitat Internacional de Catalunya, Barcelona;* [3]*Periodontology Unit, Faculty of Odontology, University of Santiago de Compostela & Medical-Surgical Dentistry (OMEQUI) Research Group, Health Research Institute of Santiago de Compostela (IDIS), Santiago de Compostela;* [4]*Department of Statistics, Universitat Internacional de Catalunya, Barcelona, Spain;* [5]*Department of Periodontology and Oral Sciences Research Group, University of Glasgow Dental School, Glasgow, UK;* [6]*Department of Experimental Dentistry and Dental Prophylaxis, Jagiellonian University, Krakow, Poland;* [7]*Institute of Cardiovascular and Medical Sciences, University of Glasgow, Glasgow, UK;* [8]*Department of Internal and Agricultural Medicine, Jagiellonian University, Krakow, Poland; and* [9]*Genetic Epidemiology, Institute of Cardiovascular Science, Faculty of Population Health Sciences, University College London, London, UK*

Abstract. Muñoz Aguilera E, Leira Y, Miró Catalina Q, Orlandi M, Czesnikiewicz-Guzik M, Guzik TJ, Hingorani AD, Nart J, D'Aiuto F (UCL Eastman Dental Institute and Hospital, University College London, London, UK; Universitat Internacional de Catalunya, Barcelona; University of Santiago de Compostela & Medical-Surgical Dentistry (OMEQUI) Research Group, Health Research Institute of Santiago de Compostela (IDIS), Santiago de Compostela; Universitat Internacional de Catalunya, Barcelona, Spain; University of Glasgow Dental School, Glasgow, UK; Jagiellonian University, Krakow, Poland; University of Glasgow, Glasgow, UK; Jagiellonian University, Krakow, Poland; University College London, London, UK). Is systemic inflammation a missing link between periodontitis and hypertension? Results from two large population-based surveys. *J Intern Med* 2020; https://doi.org/10.1111/joim.13180

Objective. The primary objective was to investigate the relationship between periodontitis and hypertension in two independent large surveys. The secondary objective was to ascertain whether systemic inflammation had a mediation effect in the association.

Methods. This cross-sectional study analysed representative samples of the US (n = 3460; NHANES 2009/10) and Korean (n = 4539; 2015 KNHANES VI-3) populations. The association between periodontitis (exposure), hypertension (outcome) and inflammatory markers [C-reactive protein (CRP) and white blood cell counts (WBC)] (mediators) was assessed using multivariate linear and logistic regression models and mediation analysis.

Results. Participants with periodontitis were more likely to have hypertension (NHANES: OR = 1.3, 95% CI: 1.0–1.6, P = 0.025; KNHANES: OR = 1.2, 95% CI: 1.0–1.4, P = 0.041) and actual systolic blood pressure ≥ 140 mmHg (NHANES: OR = 1.6, 95% CI: 1.1–2.3, P < 0.001; KNHANES: OR = 1.3, 95% CI : 1.0–1.6, P < 0.031) than those without the disease. These associations were independent of age, gender, BMI, education level, smoking, alcohol consumption, creatinine, physical activity, presence of other comorbidities and confirmed in participants not taking antihypertensive medications. Diagnosis of periodontitis was directly associated with WBC (in both surveys: NHANES: $\beta \pm SE = 0.3 \pm 0.1$, $P < 0.004$; KNHANES: $\beta \pm SE = 0.3 \pm 0.1$, $P < 0.001$) and with CRP levels (in one survey: NHANES: $\beta \pm SE = 0.1 \pm 0.03$, $P < 0.007$; KNHANES: $\beta \pm SE = 0.1 \pm 0.04$, $P > 0.213$). Mediation analyses confirmed that CRP acted as a mediator in the association between periodontitis and hypertension in both populations (mediated effect: NHANES: $\beta \pm SE = 0.010 \pm 0.003$, $P < 0.001$; KNHANES: $\beta \pm SE = 0.003 \pm 0.001$, P = 0.015). WBC acted as a mediator in the KNHANES (mediated effect: $\beta \pm SE = 0.004 \pm 0.001$, P = 0.004) whilst in the NHANES, its effect was dependent of CRP inclusion in the model (mediated effect WBC + CRP: $\beta \pm SE = 0.002 \pm 0.001$, P = 0.001).

Conclusions. These findings suggest that periodontitis is closely linked to hypertension and systemic inflammation is, in part, a mediator of this association.

Keywords: CRP, high blood pressure, hypertension, leucocytes, periodontitis, systemic inflammation.

†Equally first author.

What is already known about this subject?

Consistent evidence suggests a direct relationship between periodontitis and hypertension. Poor oral health is linked to greater systemic inflammation and increased odds of hypertension. A linear association between systolic blood pressure and various oral health indices confirm these findings.

What does this study add?

Periodontitis increases the odds of hypertension by 20-60% in two large populations and systemic inflammation as assessed by peripheral levels of CRP and WBC acts as a biological mediator of this association.

How might this impact on clinical practice?

Oral health promotion could result in reduced systemic inflammation and it may represent a novel nonpharmacological intervention in hypertension management and its complications.

Introduction

Hypertension is a complex multifactorial disorder. Its prevalence exceeds 31% worldwide with more than 1.13 billion people affected [1]. Elevated blood pressure (BP) is strongly linked to cardiovascular complications, increasing morbidity and mortality [2]. Experimental and observational evidence supports a prominent role of systemic inflammation both in the initiation and in the progression of hypertension [3]. The management of this condition, however, is still a challenge and it represents an increasing burden for society.

Periodontitis is one of the most common inflammatory disorders worldwide with >46% of adults in the United States diagnosed with the disease [4]. Strong evidence supports the role of a dysbiotic dental biofilm in the development of periodontitis. Further, a cluster of modifiable risk factors is shared between periodontitis and leading noncommunicable diseases (NCDs) (cardiovascular diseases, cancer, chronic respiratory diseases and diabetes) [5].

Patients with periodontitis exhibit not only gingival inflammation but also endothelial dysfunction, increased bacterial burden (endotoxins and exotoxins dissemination), metabolic dysregulation and systemic inflammation [6-9]. A bidirectional link has been proposed between periodontitis and other metabolic disorders such as metabolic syndrome and diabetes [10, 11].

Hypertension has been linked to periodontitis but evidence from intervention trials is limited [12]. A possible causal relationship between these two conditions has been proposed recently using Mendelian randomization. This analysis confirmed a link between genetic variants linked to periodontitis and elevated BP phenotypes in a large UK population study [13]. The exact mechanisms mediating this association remain unknown raising the question of whether inflammation or bacterial burden could play a prominent role. Given that systemic inflammatory biomarkers such as C-reactive protein (CRP) and leucocyte counts have been correlated with both periodontitis and hypertension, we hypothesized that systemic inflammation could be a mediator between the two diseases. Therefore, confirmation of this association in large independent studies with a focus on mediators needs to be unravelled prior to undertaking interventional trials investigating the treatment of periodontitis as a target nonpharmacological treatment for hypertension. Accordingly, the primary aim of this study was to investigate the association between periodontitis and hypertension using two representative surveys of the US and Korean populations. The secondary aim was to ascertain the role of systemic inflammation in mediating this association.

Material and methods

Two population-based surveys were analysed and hereby reported in compliance with the Strengthening the Reporting of Observational studies in Epidemiology (STROBE) guidelines (Supplemental checklist).

Survey designs and study populations

Databases obtained from the US [2009/2010 National Health And Nutrition Examination Survey (NHANES)] and Korean [2015 VI-3 Korean National Health And Nutrition Examination Survey (KNHANES)] open repositories shared similar study design (stratified, multistage cluster probability sampling survey) [14, 15], and they were conducted by the statistical division of the National Centers for Disease Control and Prevention in the United States and Korea, respectively. The study

was conducted in accordance with the 1975 Declaration of Helsinki and participants provided written consent. These survey waves were selected for this study as were the first ones containing a detailed periodontal examination, measurements of average BP, serum concentrations of high-sensitivity CRP (hs-CRP) and white blood cell counts (WBC).

Exclusion criteria used in the final sample analysis were (i) age < 30 years in the NHANES ($N = 6451$) and age < 19 years in the KNHANES ($N = 1345$) as no periodontal data were collected for younger individuals; (ii) pregnancy (NHANES, $N = 23$; KNHANES, $N = 29$); (iii) lack of data on hs-CRP (NHANES, $N = 137$; KNHANES, $N = 243$); (iv) lack of data on BP (NHANES, $N = 123$; KNHANES, $N = 29$); and (v) lack of periodontal data for any other reasons (NHANES, $N = 343$, KNHANES, $N = 792$). From a total of 10 537 participants in the NHANES 2009/2010 and 6977 in the KNHANES VI-3, the final samples included in this analysis were of 3460 and 4539 participants, respectively. These populations refer to a representative sample of just over 128 millions of US and 33 millions of Korean citizens.

We extracted data on socio-demographic, healthy lifestyle behavioural factors, anthropometric measurements, medical history, oral examination, mean BP and biochemical parameters (Table S1).

Blood pressure measurements

In both cross-sectional studies, sitting BP was measured using a standardized protocol [16]. Average measurements of systolic and diastolic arterial pressure (SBP and DBP) were obtained from three consecutive readings. Participants were then categorized as normal, prehypertensive and hypertensive according to the Joint National Committee 7 guidelines [17]. Further, hypertension was defined as values of SBP $\geq$ 140 mmHg or DBP $\geq$ 90 mmHg or the use of antihypertensive medication [18]. The number of participants taking antihypertensive medications was also calculated.

Periodontal examination and dental exposure variables

The analysis was conducted using both established case definitions and continuous measures (full-mouth indices) of periodontitis. However, different protocols were used. In the NHANES, a full-mouth periodontal assessment was carried out at six sites per tooth and periodontitis (exposure) was defined as mild, moderate or severe [19]. Continuous aggregate dental variables (number and percentage of sites) were then created to indicate (a) the extent of periodontal lesions with probing pocket depths (PPD) of $\geq$4 mm, $\geq$5 mm, $\geq$6 mm and (b) the extent of loss of periodontal tissue attachment (AL) of $\geq$3 mm, $\geq$4 mm, $\geq$5 mm, $\geq$6 mm as previously described [20].

In the KNHANES study, participants presenting with community periodontal index (CPI) scores of 3 and 4 (at least in one sextant) were defined as having worse periodontal status, whereas those presenting scores of 0, 1 and 2 represented controls with better periodontal status. Continuous measures of periodontal lesions were then created as follows: (a) CPI cumulative score (the sum of only CPI scores of 3 or 4 of all sextants) and (b) CPI continuous score (sum of all CPI scores of all sextants) as previously described [21].

Laboratory analysis

Biochemical parameters were retrieved from both surveys including plasma glucose (mg dL^{-1}), insulin levels (uIU mL^{-1}), glycated haemoglobin [HbA1c (%)], total cholesterol (mg dL^{-1}), high- and low-density lipoprotein cholesterol levels [HDL and LDL (mg dL^{-1})], triglyceride levels (mg dL^{-1}), creatinine (mg dL^{-1}), hs-CRP (mg dL^{-1}) and WBCs (thous μL^{-1})].

Statistical analysis

Data analyses were performed with STATA version 15.0 (StataCorp, College Station, Tex, USA) and R Software (version 3.5.2). Continuous variables are reported as mean $\pm$ standard error (SE), whereas categorical variables are expressed as percentages. Simple differences between participants with or without periodontitis were assessed by independent t-test (for continuous variables) or chi-square test (for categorical variables). Normality assumptions were checked, and a logarithmic transformation of hs-CRP was used for parametric analyses. Different measures of oral disease exposure (categorical and continuous) were adopted to test the association between periodontal status and BP. Further, circulating levels of WBC and hs-CRP were used as biomarkers of systemic inflammation and possible mediators of the association between periodontal status and hypertension. Univariate analyses were performed for all continuous

variables comparing the groups of participants with periodontitis/worse periodontal status and the rest of the study sample. All those variables with statistically significant associations were then used in the multivariate models.

Multivariate logistic regression models were created to test potential associations between periodontitis case definitions or continuous measures of periodontal lesions with hypertension, SBP ≥ 140 mmHg or hs-CRP > 2mg L^{-1} as outcome variables. Multivariate linear regression models were then constructed to investigate the association between periodontal (both categorical and continuous) and arterial BP (mean SBP/DBP) variables. Similar analyses were performed with hs-CRP and WBC values (exposures) and hypertension (as categorical or continuous outcomes). Odds ratios (ORs) and 95% confidence intervals (CI) were calculated as well as β coefficient with standard errors. A fully adjusted model (Model 1) included age, gender, ethnicity, smoking, education level and chronic medical conditions as covariates (as previously reported) in both surveys [18]. In addition to these, body mass index (BMI), alcohol consumption, creatinine and physical activity were included in the multivariable models of the KNHANES survey (as they all presented univariate association with the outcome variables).

Sensitivity analyses in the subgroup of participants not taking antihypertensive medications were also performed (Model 2) (NHANES, $N = 2486$; KNHANES, $N = 3270$).

Structural equation modelling (SEM) was then used to estimate whether the association between periodontitis and hypertension was mediated by WBC or CRP using R Software [22]. Four different and prespecified routes were used as follows: direct (route 1) and indirect (route 2, 3, 4) mediation effects with their 95% CI were estimated:

Route 1: Periodontitis (exposure) → Hypertension (outcome).

Route 2: Periodontitis (exposure) → WBC (mediator) → Hypertension (outcome).

Route 3: Periodontitis (exposure) → Log CRP (mediator) → Hypertension (outcome).

Route 4: Periodontitis (exposure) → WBC (mediator) → Log CRP (mediator) → Hypertension (outcome).

Results

Characteristics of study populations

Participants with periodontitis were predominantly men (NHANES, 60%; KNHANES, 57%), older than 50 years of age, increased number of current smokers (NHANES, 52% vs 36%; KNHANES, 23% vs 15%), of lower education background and higher prevalence of diabetes (NHANES, 12% vs 6%; KNHANES, 10% vs 5%) than participants without periodontitis (Table 1). Almost a doubled prevalence of hypertension (NHANES, 42% vs 25%; KNHANES, 39% vs 19%) and antihypertensive medication (NHANES, 31% vs 19%; KNHANES, 25% vs 12%) were observed in patients with periodontitis. Similarly, participants with periodontitis had higher values of SBP (6.4 mmHg higher in NHANES and 7.3 mmHg higher in KNHANES) than survey participants without periodontitis. In the NHANES survey, Mexican and non-Hispanic black presented with the greatest prevalence of periodontitis. Lastly, when other traditional cardiovascular risk factors were assessed, patients with periodontitis exhibited greater values of glucose, triglycerides, hs-CRP and WBC in both surveys when compared to those without periodontitis, with BMI being higher in periodontitis patients only in KNHANES (all $P < 0.001$).

Logistic regression analyses

Multiple logistic regression models confirmed that amongst participants with periodontitis and worse periodontal status, the adjusted odds of hypertension were 1.3 (95%CI 1.0–1.6) in the NHANES and 1.2 (95%CI 1.0–1.4) in the KNHANES populations, respectively (Table 2). Greater odds of hypertension in patients with periodontitis and worse periodontal status (CPI 3-4) were observed in the subgroup of participants not taking antihypertensive medications (NHANES: OR = 1.4, 95%CI 1.0–1.8, $N = 2486$; KNHANES: OR = 1.3, 95%CI 0.9–1.7, $N = 3270$). Similar associations were found between diagnosis of periodontitis and worse periodontal status (CPI 3-4) and SBP ≥ 140 values in both populations (NHANES: OR = 1.6, 95%CI 1.2–2.1; KNHANES: OR = 1.3, 95%CI 1.0–1.6) with greater odds in participants with more severe periodontitis. These findings were consistent in those participants not taking antihypertensive medications (Model 2) although the estimates were smaller than those observed in the whole sample

Table 1. *Baseline characteristics of Survey participants according to Periodontal variables*

	NHANES (2009–2010)				KNHANES VI-3 (2015)			
Variables	Overall (3460)	No-Periodontitis (1799)	Periodontitis (1661)	*P*	Overall (4539)	CPI 0-2 (2996)	CPI 3-4 (1543)	*P*
Categorical % (No.)								
Gender (% female)	49 (1695)	59 (1061)	40 (664)	**<0.0001**	43 (1952)	53 (1588)	43 (664)	**<0.0001**
Smoking	43 (1488)	36 (648)	52 (864)	**<0.0001**	17 (772)	15 (449)	23 (355)	**<0.0001**
Alcohol use	28 (969)	28 (504)	29 (482)	0.5891	25 (1335)	26 (779)	22 (339)	**0.002**
Education level				**<0.0001**				**<0.0001**
School grade	6 (208)	3 (54)	9 (150)		15 (681)	11 (330)	23 (355)	
Primary school graduate	11 (380)	8 (144)	15 (249)		9 (409)	7 (210)	14 (216)	
High school graduate	22 (761)	20 (340)	24 (399)		37 (1679)	38 (1138)	35 (540)	
College or higher	60 (2076)	69 (1241)	51 (847)		39 (1770)	44 (1318)	28 (432)	
Ethnicity				**0.0021**				
Mexican American	8 (277)	5 (90)	11 (183)					
Other Hispanics	5 (175)	5 (90)	5 (83)					
Non-Hispanics white	71 (2457)	77 (1385)	64(1063)					
Non-Hispanic black	10 (346)	8 (144)	13 (216)					
Other	6 (208)	5 (90)	7 (116)					
Diabetes	9 (311)	6 (108)	12 (199)	**0.0004**	6 (272)	5 (150)	10 (154)	**<0.00001**
Hypertension	33 (1142)	25 (450)	42 (698)	**<0.0001**	25 (1135)	19 (569)	39 (602)	**<0.0001**
Normal BP	40 (1384)	48 (864)	31 (515)		51 (2315)	56 (1678)	38 (586)	
Prehypertension	27 (934)	27 (486)	28 (465)		24 (1089)	25 (749)	23 (355)	
Antihypertension medication	25 (865)	19 (342)	31 (515)	**<0.0001**	16 (726)	12 (360)	25 (386)	**<0.0001**
Mean SBP ≥ 140 mmHg	13 (450)	8 (144)	18 (299)	**<0.0001**	9 (409)	7 (210)	15 (231)	**<0.0001**
Chronic medical conditions	52 (1799)	48 (864)	56 (930)	**0.0021**	6 (272)	5 (150)	9 (139)	**<0.0001**

Table 1 (*Continued*)

Variables	NHANES (2009–2010) Overall (3460)	No-Periodontitis (1799)	Periodontitis (1661)	*P*	KNHANES VI-3 (2015) Overall (4539)	CPI 0-2 (2996)	CPI 3-4 (1543)	*P*
Continuous (mean ± SE)								
Age (years)	51 ± 0.4	47 ± 0.4	55 ± 0.5	**<0.0001**	45.9 ± 0.4	42.3 ± 0.4	54.2 ± 0.6	**<0.0001**
BMI (kg m^{-2})	29 ± 0.1	28.8 ± 0.2	29.3 ± 0.2	0.067	23.9 ± 0.1	23.6 ± 0.1	24.6 ± 0.1	**<0.0001**
SBP (mmHg)	121.6 ± 0.5	118.5 ± 0.4	124.9 ± 0.5	**<0.0001**	117.1 ± 0.3	114.9 ± 0.3	122.2 ± 0.6	**<0.0001**
DBP (mmHg)	71.0 ± 0.6	71.4 ± 0.6	70.6 ± 0.7	**0.043**	75.5 ± 0.2	74.7 ± 0.3	77.2 ± 0.3	**<0.0001**
Physical activity[a]	3.4 ± 0.1	3.3 ± 0.1	3.5 ± 0.1	0.182	7.6 ± 0.03	7.6 ± 0.03	7.7 ± 0.04	**0.015**
Glucose (mg dL^{-1}) (NHANES: $N = 1690$)	104.2 ± 0.9	102.4 ± 1.4	108.2 ± 1.0	**<0.001**	99.6 ± 0.5	96.5 ± 0.4	106.8 ± 1.0	**<0.0001**
Insulin (uIU mL^{-1})	13.5 ± 0.3	12.8 ± 0.4	14.3 ± 0.5	**0.028**	8.5 ± 0.1	8.3 ± 0.2	8.8 ± 0.3	0.181
HbA1C (%)	5.7 ± 0.01	5.6 ± 0.01	5.8 ± 0.01	**<0.0001**	5.6 ± 0.02	5.5 ± 0.1	5.8 ± 0.1	**<0.0001**
Total cholesterol (mg dL^{-1})	201.7 ± 1.2	202.4 ± 1.3	200.9 ± 1.4	0.292	189.8 ± 0.6	188.3 ± 0.7	193.3 ± 1.1	**<0.0001**
HDL (mg dL^{-1})	53.3 ± 0.5	54.9 ± 0.6	51.6 ± 0.6	**<0.0001**	51.2 ± 0.2	54.5 ± 0.3	48.3 ± 0.4	**<0.0001**
LDL (mg dL^{-1}) (NHANES: $N = 1654$)	119.8 ± 1.1	119.7 ± 1.6	120.0 ± 1.1	0.895	113.4 ± 0.6	112.3 ± 0.6	115.8 ± 1.0	**0.003**
Triglycerides (mg dL^{-1}) (NHANES: $N = 1683$)	129.5 ± 2.3	120.7 ± 3.6	138.9 ± 3.1	**<0.0001**	138.8 ± 2.4	127.7 ± 2.6	164.7 ± 4.6	**0.0001**
Hs-CRP(mg L^{-1})	1.7 ± 1.01	1.5 ± 1.11	1.8 ± 1.01	**<0.0001**	1.2 ± 0.04	1.1 ± 0.04	1.3 ± 0.07	**0.005**
WBC (thous μL^{-1})	7.0 ± 0.04	6.9 ± 0.1	7.1 ± 0.1	**0.004**	6.5 ± 0.03	6.4 ± 0.03	6.8 ± 0.1	**<0.0001**
Creatinine[b]	118.5 ± 1.2	120.2 ± 1.9	116.7 ± 1.4	**0.113**	0.85 ± 0.01	0.8 ± 0.01	0.9 ± 0.02	**0.031**

BMI, body mass index; BP, blood pressure; DBP, diastolic blood pressure; HbA1c, glycohaemoglobin A1c; HDL, high-density lipoprotein; Hs-CRP, high-sensitivity C-reactive protein; KNHANES, Korea National Health and Nutrition Examination Survey; LDL, low-density lipoprotein; NHANES, National Health and Nutrition Examination Survey; SBP, systolic blood pressure; WBC, white blood cell counts. Bold values highlighting statistically significant results (i.e. $P < 0.05$).

[a]Physical activity *(NHANES = days/week); (KNHANES = days/month).

[b]Creatinine *(NHANES: Urine (μmol L^{-1}); KNHANES: serum mg dL^{-1}.

Table 2. *Multiple logistic regression models of hypertension and SBP ≥ 140 mmHg according to periodontal variables or systemic inflammation (hs-CRP/ WBC levels)*

		Hypertension		SBP ≥ 140 mmHg	
Survey	Exposure: Periodontal variables	Model 1 OR (95% CI)	Model 2 OR (95% CI)	Model 1 OR (95% CI)	Model 2 OR (95% CI)
NHANES	Periodontitis	**1.3 (1.0–1.6)***	**1.4 (1.0–1.8)***	**1.6 (1.2–2.1)****	**1.6 (1.1–2.3)***
	Mild	1.4 (0.8–2.2)	1.1 (0.5–2.6)	1.1 (0.6–2.0)	1.1 (0.4–2.8)
	Moderate	1.2 (1.0–1.6)	1.4 (1.0–2.0)	**1.5 (1.1–2.0)***	**1.6 (1.0–2.4)***
	Severe	1.3 (0.9–1.7)	**1.6 (1.2–2.4)****	**2.5 (1.7–3.6)*****	**2.3 (1.4–3.6)****
KNHANES	CPI 3–4 vs CPI 0–2	**1.2 (1.0–1.4)***	1.3 (0.9–1.7)	**1.3 (1.0–1.6)***	**1.4 (1.0–1.9)***
	CPI continuous	1.03 (0.9–1.1)	1.1 (0.9–1.2)	**1.1 (1.0–1.2)***	**1.1 (1.0–1.2)***
	CPI cumulative	0.9 (0.9–1.0)	1.0 (0.9–1.0)	1.0 (0.9–1.0)	1.0 (0.9–1.0)
	Exposure: Systemic inflammation				
NHANES	Hs-CRP (log)	**1.4 (1.3–1.5)*****	**1.3 (1.1–1.4)****	**1.2 (1.1–1.3)****	1.2 (1.0–1.4)
	WBC	**1.1 (1.0–1.1)****	1.1 (1.0–1.2)	1.0 (1.0–1.1)	1.0 (0.9–1.1)
KNHANES	Hs-CRP (log)	1.1 (1.0–1.2)	1.1 (0.9–1.3)	1.1 (0.9–1.2)	1.0 (0.9–1.2)
	WBC	**1.1 (1.02–1.1)****	**1.1 (1.01–1.2)***	1.0(1.0–1.1)	1.0 (0.9–1.1)

CI, confidence interval; CPI, Community Periodontal Index; Hs-CRP, high-sensitivity c-reactive protein (logarithm); KNHANES, Korea National Health and Nutrition Examination Survey; NHANES, National Health and Nutrition Examination Survey; OR, odds ratio; SBP, systolic blood pressure; WBC, white blood cell counts.
Model 1 (population sample: 3460 NHANES; 4539 KNHANES): (NHANES): Age, gender, ethnicity, smoking, education level, chronic medical condition; (KNHANES): age, gender, BMI, education level, smoking, diabetes, alcohol consumption, creatinine, physical activity, chronic medical conditions.
Model 2 (population sample: 2486 NHANES; 3270 KNHANES): Not taking antihypertensive medication.
Bold values highlighting statistically significant results.
***$P < 0.001$; **$P < 0.01$; *$P < 0.05$.

(Table 2). Indeed, NHANES participants with severe periodontitis also presented with more than twice increased likelihood of SBP ≥ 140 mmHg in model 1 (OR = 2.5 95%CI 1.7–3.6) and model 2 (OR = 2.3 95%CI 1.4–3.6), respectively (Table 2).

When hs-CRP or WBC was introduced as independent exposure variables, the odds of hypertension and SBP ≥ 140 mmHg ranged from 1.0 to 1.4 in the US and from 1.0 to 1.1 in the Korean survey. Only some continuous measurements of periodontitis (mean PPD and mean CAL) were associated with greater odds of hypertension, SBP ≥ 140 mmHg and of hs-CRP ≥ 2 mg L^{-1} in the fully adjusted model and in those participants not taking antihypertensive medications (Figure 1).

Linear regression analyses

Linear regression analyses confirmed that periodontitis (assessed both as categorical and as continuous variables) was associated with mean SBP. These findings were confirmed in the subgroup of participants not taking antihypertensive medications in the US survey (Table 3). In the Korean survey, the cumulative CPI score was consistently associated with SBP and DBP and this was also confirmed in participants not taking BP medications. Higher WBC counts were associated with mean SBP in both surveys, whilst higher hs-CRP levels were associated with SBP and DBP only in the US study (Table 3). Further, we observed a negative association between DBP and the number or percentage of gingival sites with attachment loss of ≥3 mm and of sites with probing depth ≥ 6 mm in the NHANES (model 2) and with the cumulative CPI score in the KNANES (models 1 and 2) (Table 3). Lastly, both US and Korean participants with severe periodontitis or worse periodontal status (CPI 3-4) exhibited greater systemic inflammation as assessed by hs-CRP serum levels and by WBC when compared to those without periodontitis or better periodontal status (CPI 0-2) and this difference was independent of other common confounders (Table 4).

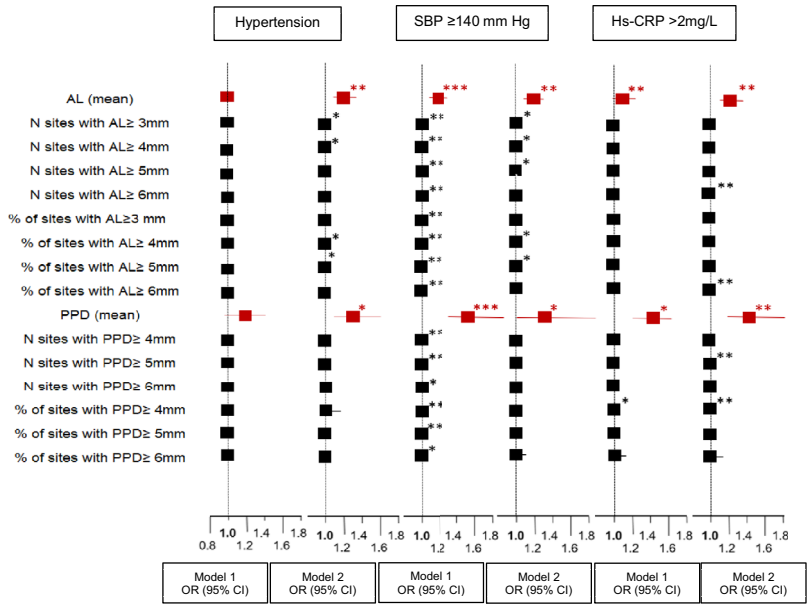


Figure 1 *Multiple logistic regression model of hypertension, SBP ≥ 140 mmHg and hs-CRP > 2 mg L^{-1} according to continuous periodontal variables (NHANES database only). NHANES, National Health and Nutrition Examination Survey; OR, odds ratio; CI, confidence interval; SBP, systolic blood pressure; hs-CRP, high-sensitivity C-reactive protein (logarithm); N, number; AL, attachment level; PPD, probing pocket depth. Model 1 (population sample: 3460 NHANES): Age, gender, ethnicity, smoking, education level, chronic medical condition. Model 2 (population sample: 2486 NHANES): Not taking antihypertensive medication.* ***P < *0.001;* **P < *0.01;* *P < *0.05.*

Mediation analyses

The association between periodontitis and hypertension (categorical) was mediated by CRP (β ± SE = 0.010 ± 0.003; $P < 0.001$) in the NHANES dataset, whilst WBC (β ± SE = 0.001 ± 0.001; $P = 0.221$) was only an indirect mediator of the association (indirect route linked to hs-CRP) (Model A, unadjusted) (Fig. 2a and Table S2). When repeating the same analysis in the KNHANES database, both hs-CRP (β ± SE = 0.003 ± 0.001; $P = 0.015$) and WBC (β ± SE = 0.004 ± 0.001; $P = 0.004$) acted as mediators of the association between worse periodontal status and hypertension (Fig. 2b and Table S2). Models B, D, C replicated these results when the analyses applied for continuous periodontal (PPD, CAL in NHANES and CPI continuous in KNANES) and BP (SBP) variables, in both adjusted and unadjusted models (Table S2).

Discussion

The analysis of two of the largest population surveys with available dental and general health data demonstrated that both categorical and continuous measures of periodontitis were consistently associated with hypertension and SBP independent of other common cardiovascular risk factors. Participants in the United States with severe periodontitis had higher odds for SBP ≥ 140 mmHg when compared to participants without periodontitis and all findings were confirmed in participants not taking antihypertensive medications. Systemic inflammation defined by two commonly measured biomarkers (hs-CRP and WBC) was not only associated independently with periodontitis, SBP and diagnosis of hypertension but acted as a modest mediator of these associations.

This analysis confirmed that participants with periodontitis have a 20-60% greater chance of presenting also a concomitant diagnosis of hypertension and a 10% to 2.5 times greater chance of SBP ≥ 140 mmHg. This is consistent with previous studies reporting that patients with periodontitis have on average 4.5 higher mean SBP (95% CI: 2.88–6.11) than participants without periodontitis [12]. In the present study, a higher mean SBP of 6.4 mmHg (NHANES, 95%CI 5.3–7.4) and of 7.2 mmHg (KNHANES, 95%CI 6.1–8.4) were observed when participants with periodontitis were

Table 3. *Linear regression models of SBP and DBP according to periodontal variables or systemic inflammation (hs-CRP or WBC levels)*

Survey	Exposure: Periodontal variables	SBP Model 1 ß ± SE	SBP Model 2 ß ± SE	DBP Model 1 ß ± SE	DBP Model 2 ß ± SE
NHANES	Case Definition (vs non perio)				
	Periodontitis	**1.7 ± 0.7***	**1.5 ± 0.7***	−0.1 ± 0.4	−0.5 ± 0.4
	Mild	−0.2 ± 1.0	−0.4 ± 1.2	0.7 ± 0.6	0.6 ± 0.7
	Moderate	**1.7 ± 0.6****	**1.6 ± 0.7***	−0.4 ± 0.4	−1.0 ± 0.5
	Severe	**4.9 ± 1.3****	**4.7 ± 1.6***	−0.2 ± 0.7	−0.5 ± 0.8
	Continuous variable				
	AL (mean)	**1.3 ± 0.4****	**1.6 ± 0.5****	−0.1 ± 0.2	−0.2 ± 0.3
	N sites with AL ≥ 3 mm	**0.1 ± 0.04****	**0.03 ± 0.01***	−0.01 ± 0.01	**−0.01 ± 0.0***
	N sites with AL ≥ 4 mm	**0.1 ± 0.1****	**0.1 ± 0.02****	−0.02 ± 0.02	−0.02 ± 0.02
	N sites with AL ≥ 5 mm	**0.1 ± 0.03***	**0.1 ± 0.03***	0.01 ± 0.01	−0.01 ± 0.01
	N sites with AL ≥ 6 mm	**0.1 ± 0.1***	**0.1 ± 0.1***	0.02 ± 0.02	0.02 ± 0.02
	% of sites with AL ≥ 3 mm	**7.7 ± 2.5****	**5.6 ± 2.0***	−1.8 ± 1.4	**−3.8 ± 1.5***
	% of sites with AL ≥ 4 mm	**11.5 ± 3.5****	**9.6 ± 2.9****	−1.7 ± 1.6	−3.1 ± 2.0
	% of sites with AL ≥ 5 mm	**16.0 ± 5.4***	**13.1 ± 5.4***	0.8 ± 2.2	−1.5 ± 2.9
	% of sites with AL ≥ 6 mm	**23.8 ± 8.5***	**19.5 ± 8.9***	4.5 ± 3.7	0.8 ± 4.5
	PPD (mean)	**2.3 ± 0.6****	1.1 ± 0.7	0.01 ± 0.4	−0.6 ± 0.4
	N sites with PPD ≥ 4 mm	**0.1 ± 0.01****	0.01 ± 0.01	0.01 ± 0.02	−0.01 ± 0.01
	N sites with PPD ≥ 5 mm	**0.2 ± 0.1****	0.1 ± 0.1	0.01 ± 0.02	−0.1 ± 0.02
	N sites with PPD ≥ 6 mm	0.3 ± 0.1	0.1 ± 0.1	−0.01 ± 0.02	**−0.1 ± 0.02***
	% of sites with PPD ≥ 4 mm	**16.3 ± 4.0****	7.0 ± 3.7	0.9 ± 2.9	−4.4 ± 2.3
	% of sites with PPD ≥ 5 mm	**32.0 ± 9.1****	**17.6 ± 8.5***	0.7 ± 4.7	−8.6 ± 4.5
	% of sites with PPD ≥ 6 mm	44.2 ± 22.6	21.3 ± 21.7	−1.5 ± 5.7	**−16.5 ± 6.7***
KNHANES	CPI 3–4 vs CPI 0–2	0.7 ± 0.5	0.4 ± 0.6	0.7 ± 0.4	0.2 ± 0.5
	CPI continuous	0.3 ± 0.2	0.2 ± 0.2	0.2 ± 0.1	0.1 ± 0.1
	CPI cumulative	**0.1 ± 0.03***	**0.1 ± 0.04***	**−0.1 ± 0.1****	**−0.1 ± 0.1***
	Exposure: Systemic Inflammation				
NHANES	hs-CRP (log)	**1.2 ± 0.2*****	**1.3 ± 0.3*****	**0.7 ± 0.2****	**0.7 ± 0.2***
	WBC	**0.5 ± 0.1****	**0.6 ± 0.2****	−0.03 ± 0.1	0.1 ± 0.2
KNHANES	hs-CRP (log)	0.4 ± 0.2	0.4 ± 0.3	0.1 ± 0.2	0.1 ± 0.2
	WBC	**0.5 ± 0.1*****	**0.7 ± 0.2*****	**0.3 ± 0.1****	**0.5 ± 0.1*****

AL, attachment loss; CPI, Community Periodontal Index; DBP, diastolic blood pressure; hs-CRP, high-sensitivity c-reactive protein (logarithm); KNHANES, Korea National Health and Nutrition Examination Survey; NHANES, National Health and Nutrition Examination Survey; PPD, probing pocket depth; SBP, systolic blood pressure; SE, standard error; WBC, white blood cell counts.

Model 1 (Population sample: 3460 NHANES; 4539 KNHANES): (NHANES): Age, gender, ethnicity, smoking, education level, chronic medical condition; (KNHANES): Age, gender, BMI, education level, smoking, diabetes, alcohol consumption, creatinine, physical activity, chronic medical conditions.

Model 2 (population sample: 2486 NHANES; 3270 KNHANES): Not taking antihypertensive medication.

Bold values highlighting statisticaly significant results.

***$P < 0.001$; **$P < 0.01$; *$P < 0.05$.

Table 4. *Linear regression model of CRP and WBC according to periodontal variables*

Survey	Exposure: Periodontal variables	Unadjusted ß ± SE	Model 1 ß ± SE
hs-CRP (log)			
NHANES	Periodontitis	**0.2 ± 0.01*****	**0.1 ± 0.03****
	Mild	**0.3 ± 0.1****	0.1 ± 0.1
	Moderate	**0.2 ± 0.1****	0.1 ± 0.02
	Severe	**0.3 ± 0.1****	**0.3 ± 0.1****
KNHANES	CPI 3-4 vs CPI 0-2	**0.3 ± 0.04*****	0.1 ± 0.04
	CPI continuous	**0.1 ± 0.01*****	**0.03 ± 0.01***
	CPI cumulative	**0.02 ± 0.01*****	**0.01 ± 0.01*****
WBC			
NHANES	Periodontitis	**0.3 ± 0.1****	**0.3 ± 0.1****
	Mild	**0.4 ± 0.1***	**0.3 ± 0.1***
	Moderate	0.1 ± 0.1	0.1 ± 0.1
	Severe	**0.8 ± 0.2****	**0.7 ± 0.1*****
KNHANES	CPI 3-4 vs CPI 0-2	**0.4 ± 0.1*****	**0.3 ± 0.1*****
	CPI continuous	**0.2 ± 0.02*****	**0.1 ± 0.03*****
	CPI cumulative	**0.02 ± 0.01*****	**0.02 ± 0.01*****

CPI, Community Periodontal Index; Hs-CRP, high-sensitivity C-reactive protein (logarithm); KNHANES, Korea National Health and Nutrition; Examination Survey; NHANES, National Health and Nutrition Examination Survey; SE, Standard Error; WBC, White Blood Cell Counts.
Model 1 (population sample: 3460 NHANES; 4539 KNHANES): (NHANES): Age, gender, ethnicity, smoking, education level, chronic medical condition; (KNHANES): Age, gender, BMI, education level, smoking, diabetes, alcohol consumption, creatinine, physical activity, chronic medical conditions.
Bold values highlighting statistically significant results.
***$P < 0.001$; **$P < 0.01$; *$P < 0.05$.

compared to those without the disease. The magnitude of this association could have important public health implications if we consider that high sodium intake with the diet is linked to a 6.0mmHg higher average SBP [23]. Further, SBP is a strong independent risk predictor for coronary heart disease events, stroke, heart failure and end-stage renal disease [24, 25].

Negligible associations of measures of periodontitis with DBP have been reported [20, 26]. Interestingly, we observed a negative linear association between two measures of continuous disease and DBP in both datasets. This observation has not been reported previously, and it seems to be in contrast with the findings related to SBP. At this stage, it is speculative to suggest a biological explanation of these findings. Authors consider important the role of residual confounding from other traditional risk factors variables (i.e. age, gender, ethnicity) as the estimates of association between DBP and some of the continuous measures of periodontitis tended to be greater in multivariate fully adjusted models. DBP is not considered on its own as a strong predictor for CVD events [27, 28], and the role of inflammation on affecting this measure of blood pressure is unclear. Further research should be conducted to ascertain the degree of association between diastolic pressure and periodontal inflammation as well as investigate potential biological mechanisms linking them.

When comparing the findings between the two surveys, a stronger association of categorical and continuous variables of periodontitis and gingival inflammation with hypertension and SBP was observed in the US dataset when compared with the Korean data survey. Similar findings were found for the association between biomarkers of inflammation (CRP and leucocytes counts) with measures of arterial blood pressure. Whilst the two sample populations present ethnic and socio-economic differences including a higher proportion of current smokers, adiposity and chronic medical

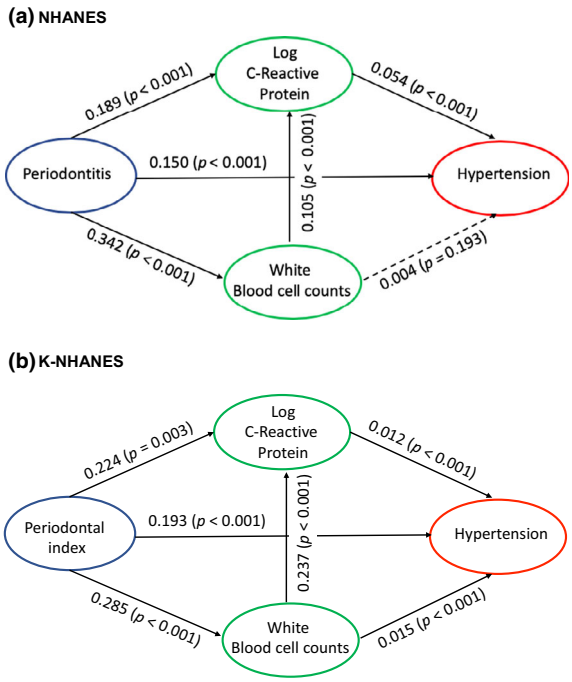


Figure 2 *Mediation analysis model. (a) NHANES. Mediation models of periodontitis, inflammation and hypertension (unadjusted) in NHANES (*N *= 3460): Route 1: Direct effect (0.150;* P *< 0.001) of periodontitis (exposure) towards hypertension (outcome). Route 2: Indirect effect (0.342;* P *< 0.001) of periodontitis (exposure) towards WBC (mediator) is shown, but no effect (0.004;* P *= 0.193) is observed in the last step of the model from WBC (mediator) towards hypertension (outcome). Route 3: Indirect effect (0.18;* P *< 0.001), of periodontitis (exposure) is observed towards Log CRP (mediator) and an effect (0.054;* P *< 0.001), from Log CRP (mediator) towards hypertension (outcome). Route 4: Indirect effect (0.342;* P *< 0.001) of periodontitis (exposure) towards WBC (mediator) and an effect (0.105;* P *< 0.001) from WBC towards Log CRP (mediator) and an effect (0.054;* P *< 0.001) from the Log CRP (mediator) towards hypertension (outcome). (b) K-NHANES. Mediation models of periodontitis, inflammation and hypertension (unadjusted) in K-NHANES (*N *= 4539): Route 1: Direct effect (0.193;* P *< 0.001) of periodontal index (exposure) towards hypertension (outcome). Route 2: Indirect effect (0.285;* P *< 0.001) of periodontal index (exposure) towards WBC (mediator), and an effect (0.015;* P *< 0.001) from WBC (mediator) towards hypertension (outcome). Route 3: Indirect effect (0.224;* P *= 0.003), of periodontal index (exposure) is observed towards Log CRP (mediator) and an effect (0.012;* P *< 0.001), from Log CRP (mediator) towards Hypertension (outcome). Route 4: Indirect effect (0.285;* P *< 0.001) of periodontal index (exposure) towards WBC (mediator) and an effect (0.237;* P *< 0.001) from WBC towards Log CRP (mediator) and an effect (0.012;* P *< 0.001) from the Log CRP (mediator) towards hypertension (outcome).*

conditions in the US population, authors believe that the different clinical measures of periodontitis recorded in the surveys could influence the results of the analyses.

A recent intervention study assessing the impact of periodontitis treatment on arterial blood pressure confirmed a substantial reduction of SBP after 2 months (mean difference of 11.1 mmHg) [13]. This preliminary evidence suggests that periodontal treatment could represent a novel nonpharmacological intervention for hypertension of similar magnitude of other lifestyles adjustments (weight loss, increasing physical activity, salt or alcohol intake reduction or smoking cessation) with an average reduction of SBP ranging from 4.6 to 6.4 mmHg [29-31]. However, larger and longer RCTs are needed.

Several lines of evidence now implicate inflammation in the development and progression of vascular diseases. For the last 3 decades, inflammation has been recognized as a common denominator of early vascular dysfunction, leading onto the development of atheroma and vascular complications [32]. Recent proof-of-concept evidence suggests that targeting upstream inflammation by selective drugs results in reduced morbidity and mortality [33]. This could also be applicable in hypertension. Experimental and human studies have documented several pathways by which elevated inflammatory markers such as CRP and circulating leucocytes are associated with an increased risk of incident hypertension including a derangement of the renin–angiotensin system, increased oxidative stress and downregulation of nitric oxide leading to increased endothelial stiffness and dysfunction [34]. A recent review identified a number of potential sources of extravascular inflammation including periodontitis as a potential factor influencing vascular risk [32]. It is now well documented that patients with periodontitis have elevated levels of CRP and WBC [7, 35].

In the mediation analysis, our findings suggest that CRP and WBC mediate partly the association between periodontitis and hypertension, although the effect is rather modest in nature (only 2% of the association explained by the model for the Korean survey whilst up to 7% in the US survey). Similar findings were recently reported for CRP (5.4%), WBC (4.2%) and ferritin (10.2%) as mediators of the total association between a continuous measure of periodontitis and high/uncontrolled BP

(≥130/80 mmHg) [36]. An alternative pathway implicated in hypertension and cardiovascular injury relates to the activation of innate and adaptive immune cells such as monocyte/macrophages, and B and T lymphocytes [37]. Damage-activated molecular patterns from the vasculature and pathogen-activated molecular patterns from opportunistic diseases such as periodontitis can exacerbate the inflammatory cascade by activation of Th1 and Th17 lymphocytes, with kidneys and vasculature injuries aggravating a pro-hypertensive status, which results in progressive raised BP [38, 39].

Our analyses point perhaps towards a more prominent role of the gut and oral microbiome and their dysbiosis on hypertension [40]. Periodontal pathogens may well play a role influencing the gut microbiome as well as exerting a direct vascular effect. Swallowing Gram-negative oral bacteria or their end-products may trigger metabolic endotoxemia and systemic inflammation contributing to cardio-metabolic disorders [41]. Lastly, experimental studies confirmed that periodontal bacteria can cause lower nitric oxide bioavailability and vascular dysfunction [42] and patients with periodontitis exhibit less nitrate-reducing bacteria [43]. These novel mechanistic hypotheses warrant further investigation.

Cross-sectional designs preclude any inference on a possible temporal and/or causal association between periodontitis and hypertension. In the attempt of mitigating this limitation, we performed the analysis in two large surveys as to identify common patterns of association and minimizing spurious findings. Two different periodontal assessments and case definitions were adopted in each survey which could be considered a limitation but could also show that the association remains significant irrespectively. Whilst in the NHANES, a recognized case definition was used [19], in the Korean survey a simplified clinical index (CPI) was selected, which is known to have risks of overestimation of the extent but underestimation of the prevalence of periodontitis [44]. In the attempt to overcome some of these limitations, we included a panel of measures of periodontal lesions to detect whether simple categorical associations were replicated when using other exposure variables. Another limitation to consider is the effect of antihypertensive medications on gingival inflammation and increased probing depths as well as on the overall association between periodontitis and hypertension [45]. Sensitivity analyses were therefore performed by repeating the models in the group of participants not taking antihypertensive medications. We cannot, however, exclude that our analyses missed some common risk determinants for hypertension (abdominal obesity, salt intake, use of anti-inflammatory drugs, hormone treatments and stress) as well as unmeasured confounders associated with both periodontitis and hypertension (residual confounding). Future mechanistic and clinical studies should investigate further the role of periodontal-driven systemic inflammation and microbial burden as a risk factor for the development and management of hypertension and its complications.

Conclusion

Periodontitis is closely linked to hypertension and low-grade systemic inflammation could be a key mediator in the association. Further interventional studies are needed to ascertain whether the treatment of periodontitis, leading to a decrease in systemic inflammation, may represent a novel nonpharmacologic intervention in hypertension management.

Acknowledgments

The study was designed and carried out by E Muñoz Aguilera, Y Leira and F D'Aiuto. These three authors together with Q Miro performed the statistical analyses and interpreted the results. E Muñoz Aguilera, Y Leira and F D'Aiuto drafted the manuscript. J Nart, M Orlandi, M Czesnikiewicz-Guzik, TJ. Guzik, AD. Hingorani provided critical interpretation and revision of the manuscript. All gave final approval and agreed to be accountable for all aspects of work ensuring integrity and accuracy.

Author contribution

Eva Maria Muñoz Aguilera: Conceptualization (equal); Data curation (equal); Formal analysis (equal); Investigation (equal); Methodology (equal); Project administration (equal); Resources (equal); Software (equal); Supervision (supporting); Validation (lead); Visualization (lead); Writing-original draft (lead); Writing-review & editing (lead). **Yago Leira Feijoo:** Conceptualization (equal); Data curation (equal); Formal analysis (equal); Investigation (equal); Methodology (equal); Project administration (equal); Resources (equal); Software (equal); Supervision (supporting); Validation (equal); Visualization (equal); Writing-original draft

(equal); Writing-review & editing (supporting). **Queralt Miró Catalina:** Conceptualization (supporting); Data curation (supporting); Formal analysis (lead); Investigation (supporting); Methodology (supporting); Resources (supporting); Software (lead); Supervision (supporting); Validation (supporting); Visualization (supporting); Writing-review & editing (supporting). **Marco Orlandi:** Conceptualization (supporting); Data curation (supporting); Formal analysis (supporting); Investigation (supporting); Methodology (supporting); Resources (equal); Software (supporting); Supervision (supporting); Validation (supporting); Visualization (supporting); Writing-original draft (supporting); Writing-review & editing (supporting). **Marta Czesnikiewicz-Guzik:** Conceptualization (supporting); Investigation (supporting); Methodology (supporting); Project administration (supporting); Resources (equal); Software (supporting); Supervision (supporting); Validation (supporting); Visualization (supporting); Writing-original draft (supporting); Writing-review & editing (supporting). **Tomasz Guzik:** Conceptualization (supporting); Formal analysis (supporting); Investigation (supporting); Methodology (supporting); Resources (supporting); Supervision (supporting); Validation (supporting); Writing-original draft (supporting); Writing-review & editing (supporting). **Aaron Hingorani:** Conceptualization (supporting); Data curation (supporting); Formal analysis (supporting); Investigation (supporting); Methodology (supporting); Supervision (supporting); Validation (supporting); Writing-original draft (supporting); Writing-review & editing (supporting). **José Nart Molina:** Conceptualization (lead); Data curation (supporting); Formal analysis (supporting); Investigation (supporting); Methodology (supporting); Project administration (lead); Resources (supporting); Software (supporting); Supervision (supporting); Validation (supporting); Visualization (supporting); Writing-original draft (supporting); Writing-review & editing (supporting). **Francesco D'Aiuto:** Conceptualization (equal); Data curation (lead); Formal analysis (lead); Funding acquisition (equal); Investigation (equal); Project administration (equal); Resources (supporting); Software (supporting); Supervision (lead); Validation (supporting); Visualization (supporting); Writing-original draft (supporting); Writing-review & editing (supporting).

Sources of funding

We would like to acknowledge that contribution of this work was undertaken at UCLH/UCL who received a proportion of funding from the Department of Health's NIHR Biomedical Research Centre funding scheme. Dr. Leira holds a Senior Clinical Research Fellowship supported by the UCL Biomedical Research Centre who receives funding from the NIHR. Marco Orlandi holds a NIHR Clinical Lectureship. Tom JGuzik is funded by European Research Council (InflammaTENSION; ERC-CoG-726318) and ERA-NET CVD (PLAQUEFIGHT; 01KL1808 to T.J.G./NCBiR, Poland).

Patient and public involvement

This research was done without patient involvement. Patients were not invited to comment on the study design and were not consulted to develop patient-relevant outcomes or interpret the results. Patients were not invited to contribute to the writing or editing of this document for readability or accuracy.

Conflict of interest statement

None.

References

1 Zhou B, Bentham J, Di Cesare M *et al.* Worldwide trends in blood pressure from 1975 to 2015: a pooled analysis of 1479 population-based measurement studies with 19·1 million participants. *Lancet* 2017; **389:** 37–55.
2 Kirkland EB, Heincelman M, Bishu KG *et al.* Trends in healthcare expenditures among US adults with hypertension: national estimates, 2003–2014. *J Am Heart Assoc* 2018; **7:** e008731.
3 Drummond GR, Vinh A, Guzik TJ, Sobey CG. Immune mechanisms of hypertension. *Nat Rev Immunol* 2019; **19:** 517–32.
4 Eke PI, Dye BA, Wei L *et al.* Update on prevalence of periodontitis in adults in the United States: NHANES 2009 to 2012. *J Periodontol* 2015; **86:** 611–22.
5 Sanz M, Marco del Castillo A, Jepsen S *et al.* Periodontitis and cardiovascular diseases: Consensus report. *J Clin Periodontol* 2020; **47:** 268–88.
6 Desvarieux M, Demmer RT, Jacobs DR Jr *et al.* Periodontal bacteria and hypertension: the oral infections and vascular disease epidemiology study (INVEST). *J Hypertens* 2010; **28:** 1413.
7 Paraskevas S, Huizinga JD, Loos BG. A systematic review and meta-analyses on C-reactive protein in relation to periodontitis. *J Clin Periodontol* 2008; **35:** 277–90.
8 Orlandi M, Suvan J, Petrie A *et al.* Association between periodontal disease and its treatment, flow-mediated dilatation and carotid intima-media thickness: a systematic review and meta-analysis. *Atherosclerosis* 2014; **236:** 39–46.
9 Tonetti MS, Dyke TE. Periodontitis and atherosclerotic cardiovascular disease: consensus report of the Joint EFP/AAP

Workshop on Periodontitis and Systemic Diseases. *J Clin Periodontol* 2013; **40:** S24–S29.
10 Preshaw P, Alba A, Herrera D *et al.* Periodontitis and diabetes: a two-way relationship. *Diabetologia* 2012; **55:** 21–31.
11 Jepsen S, Suvan J, Deschner J. The association of periodontal diseases with metabolic syndrome and obesity. *Periodontol 2000.* 2020; **83:** 125–53.
12 Muñoz Aguilera E, Suvan J, Buti J *et al.* Periodontitis is associated with hypertension: a systematic review and meta-analysis. *Cardiovasc Res* 2020; **116:** 28–39.
13 Czesnikiewicz-Guzik M, Osmenda G, Siedlinski M *et al.* Causal association between periodontitis and hypertension: evidence from Mendelian randomization and a randomized controlled trial of non-surgical periodontal therapy. *Eur Heart J* 2019; **40:** 3459–70.
14 Johnson CL, Paulose-Ram R, Ogden CL *et al. National health and nutrition examination survey. Analytic guidelines, 1999–2010.* 2013.
15 Kweon S, Kim Y, Jang M-j *et al.* Data resource profile: the Korea national health and nutrition examination survey (KNHANES). *Int J Epidemiol* 2014; **43:** 69–77.
16 Ostchega Y, Prineas RJ, Paulose-Ram R, Grim CM, Willard G, Collins D. National Health and Nutrition Examination Survey 1999–2000: effect of observer training and protocol standardization on reducing blood pressure measurement error. *J Clin Epidemiol* 2003; **56:** 768–74.
17 Chobanian AV, Bakris GL, Black HR *et al.* The seventh report of the joint national committee on prevention, detection, evaluation, and treatment of high blood pressure: the JNC 7 report. *JAMA* 2003; **289:** 2560–71.
18 Mills KT, Bundy JD, Kelly TN *et al.* Global disparities of hypertension prevalence and control: a systematic analysis of population-based studies from 90 countries. *Circulation* 2016; **134:** 441–50.
19 Eke PI, Page RC, Wei L, Thornton-Evans G, Genco RJ. Update of the case definitions for population-based surveillance of periodontitis. *J Periodontol* 2012; **83:** 1449–54.
20 Tsakos G, Sabbah W, Hingorani AD *et al.* Is periodontal inflammation associated with raised blood pressure? Evidence from a National US survey. *J Hypertens* 2010; **28:** 2386–93.
21 Masi S, Gkranias N, Li K *et al.* Association between short leukocyte telomere length, endotoxemia, and severe periodontitis in people with diabetes: a cross-sectional survey. *Diabetes Care* 2014; **37:** 1140–7.
22 Team RC. *R: A Language and Environment for Statistical Computing.* R Foundation for Statistical Computing: Vienna. https.www.R-project.org, 2017.
23 Elliott P, Stamler J, Nichols R *et al.* Intersalt revisited: further analyses of 24 hour sodium excretion and blood pressure within and across populations. *BMJ* 1996; **312:** 1249–53.
24 Collins R, Peto R, MacMahon S *et al.* Blood pressure, stroke, and coronary heart disease: part 2, short-term reductions in blood pressure: overview of randomised drug trials in their epidemiological context. *Lancet* 1990; **335:** 827–38.
25 Stamler J, Stamler R, Neaton JD. Blood pressure, systolic and diastolic, and cardiovascular risks: US population data. *Arch Intern Med* 1993; **153:** 598–615.
26 Pietropaoli D, Del Pinto R, Ferri C *et al.* Poor oral health and blood pressure control among US hypertensive adults: results from the National Health and Nutrition Examination Survey 2009 to 2014. *Hypertension* 2018; **72:** 1365–73.
27 Kannel WB, Gordon T, Schwartz MJ. Systolic versus diastolic blood pressure and risk of coronary heart disease: the Framingham study. *Am J Cardiol* 1971; **27:** 335–46.
28 Domanski M, Mitchell G, Pfeffer M *et al.* Pulse pressure and cardiovascular disease–related mortality: follow-up study of the Multiple Risk Factor Intervention Trial (MRFIT). *JAMA* 2002; **287:** 2677–83.
29 Sacks FM, Svetkey LP, Vollmer WM *et al.* Effects on blood pressure of reduced dietary sodium and the Dietary Approaches to Stop Hypertension (DASH) diet. *N Engl J Med* 2001; **344:** 3–10.
30 Neter JE, Stam BE, Kok FJ, Grobbee DE, Geleijnse JM. Influence of weight reduction on blood pressure: a meta-analysis of randomized controlled trials. *Hypertension* 2003; **42:** 878–84.
31 Whelton SP, Chin A, Xin X, He J. Effect of aerobic exercise on blood pressure: a meta-analysis of randomized, controlled trials. *Ann Intern Med* 2002; **136:** 493–503.
32 Libby P, Loscalzo J, Ridker PM *et al.* Inflammation, immunity, and infection in atherothrombosis: JACC review topic of the week. *J Am Coll Cardiol* 2018; **72:** 2071–81.
33 Ridker PM, MacFadyen JG, Everett BM *et al.* Relationship of C-reactive protein reduction to cardiovascular event reduction following treatment with canakinumab: a secondary analysis from the CANTOS randomised controlled trial. *Lancet* 2018; **391:** 319–28.
34 Guzik TJ, Touyz RM. Oxidative stress, inflammation, and vascular aging in hypertension. *Hypertension* 2017; **70:** 660–7.
35 Loos BG. Systemic markers of inflammation in periodontitis. *J Periodontol* 2005; **76:** 2106–15.
36 Pietropaoli D, Del Pinto R, Ferri C *et al.* Association between periodontal inflammation and hypertension using periodontal inflamed surface area and bleeding on probing. *J Clin Periodontol* 2020; **47:** 160–72.
37 Guzik TJ, Hoch NE, Brown KA *et al.* Role of the T cell in the genesis of angiotensin II–induced hypertension and vascular dysfunction. *J Exp Med* 2007; **204:** 2449–60.
38 Krishnan SM, Ling YH, Huuskes BM *et al.* Pharmacological inhibition of the NLRP3 inflammasome reduces blood pressure, renal damage, and dysfunction in salt-sensitive hypertension. *Cardiovasc Res* 2019; **115:** 776–87.
39 Wang H-X, Li W-J, Hou C-L *et al.* CD1d-dependent natural killer T cells attenuate angiotensin II-induced cardiac remodelling via IL-10 signalling in mice. *Cardiovasc Res* 2019; **115:** 83–93.
40 Santisteban MM, Qi Y, Zubcevic J *et al.* Hypertension-linked pathophysiological alterations in the gut. *Circ Res* 2017; **120:** 312–23.
41 Arimatsu K, Yamada H, Miyazawa H *et al.* Oral pathobiont induces systemic inflammation and metabolic changes associated with alteration of gut microbiota. *Sci Rep* 2014; **4:** 4828.
42 Velsko IM, Chukkapalli SS, Rivera MF *et al.* Active invasion of oral and aortic tissues by Porphyromonas gingivalis in mice causally links periodontitis and atherosclerosis. *PLoS One* 2014; **9:** e97811.
43 Ashworth A, Cutler C, Farnham G *et al.* Dietary intake of inorganic nitrate in vegetarians and omnivores and its impact on blood pressure, resting metabolic rate and the oral microbiome. *Free Radic Biol Med* 2019; **138:** 63–72.

44 Tran DT, Gay I, Du XL *et al.* Assessment of partial-mouth periodontal examination protocols for periodontitis surveillance. *J Clin Periodontol* 2014; **41:** 846–52.
45 Rodrigues M, Barbirato D, Luiz R, Scharfstein J, Salles G, Feres-Filho E. Effect of antihypertensive therapy with angiotensin-converting enzyme inhibitors on chronic periodontitis: a case–control study. *Oral Dis.* 2016; **22:** 791–6.

Correspondence: Francesco D'Aiuto, Periodontology Department, UCL Eastman Dental Institute 21 University Street, London WC1E 6DE, UK.
(e-mail: f.daiuto@ucl.ac.uk).

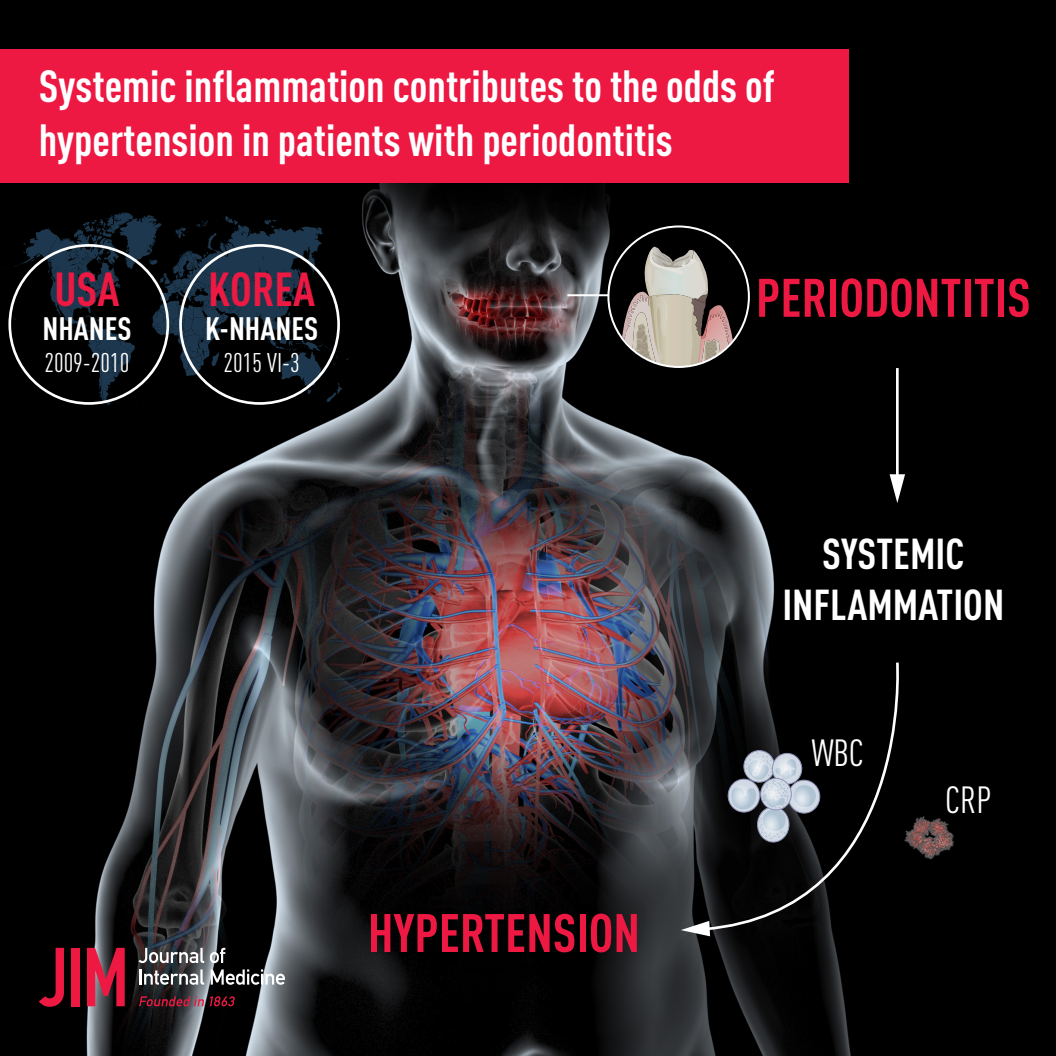
Supporting Information

Additional Supporting Information may be found in the online version of this article:

Table S1. Variables description for Korean and American databases.

Table S2. (Models A, B, C, D): Mediation analyses.

Systemic inflammation contributes to the odds of hypertension in patients with periodontitis



JIM Journal of Internal Medicine
Founded in 1863

Table S1: Variables description for Korean and American databases

VARIABLES		KOREAN (DESCRIPTION)	METHOD	AMERICAN (DESCRIPTION)	METHOD
Socio-demographic	**Age (ODV)**	Years old upon examination	Examination	Years old upon examination	Examination
	Gender (ODV)	(female/male)	Examination	(female/male)	Examination
	Education level (MDV in NHANES to match KNHANES description)	Below graduate	Questionnaire	<9th grade	Questionnaire
		Primary school graduate		9th-11th grade	
		High school graduate		High school	
		College or higher		College or above, some college or AA degree	
Health lifestyle behaviour	**Smoking (NCV)**	Smokers: Currently smoking and smoking >100 cigarettes in their whole life.	Questionnaire	Smokers: Currently smoking and smoking >100 cigarettes in their whole life .	Questionnaire
	Alcohol consumption (NCV)	Alcohol consumers: Participants that have drunk alcohol ≥2 times/week in one year	Questionnaire	Alcohol consumers: Participants that have drunk alcohol ≥2 times/week in one year	Questionnaire
	Physical activity (NCV)	Middle strength = if 1-9 days per month	Questionnaire	Middle strength= if 2 days per week/1-9 per month	Questionnaire
Anthropo-metric measurement	**BMI (ODV)**	Calculated using the formula weight divided by the square of height (Kg/m^2)	Measured following standardized protocols	Calculated using the formula weight divided by the square of height (Kg/m^2)	Measured following standardized protocols
Systemic health	**Chronic conditions (NCV)**	At least one of the following included: stroke, myocardial infarction, angina, rheumatoid arthritis, kidney failure and asthma	Questionnaire	At least one of the following included: diabetes, asthma, psoriasis, gout, congestive heart failure, coronary heart disease, angina heart attack, stroke, emphysema, thyroid, bronchitis, liver and cancer.	Questionnaire
	Diabetes (ODV)	Fasting plasma glucose ≥126 mg/dL, or medical diagnosis of diabetes by a trained medical professional, or medicated for diabetes (hypoglycaemic agents or insulin injections)	Laboratory analysis, medical diagnose or questionnaire	Presence or absence in Questionnaire	Questionnaire
BP measurement	**SBP (mmHg) (ODV)**	3 consecutive BP measurements after 6 min resting time and in the sitting	Examination	3 consecutive BP measurements in the sitting position were obtained by	Examination

	DBP (mmHg) (ODV)	position were obtained by trained professionals		trained professionals	
Hypertensive status	**Hypertension (ODV in KHANES, NCV in NHANES)**	SBP ≥140 mmHg or DBP ≥90 mmHg or taking antihypertensives	Defined according to JNC 7 (Chobanian et al. 2004)	SBP ≥140 mmHg or DBP ≥90 mmHg or taking antihypertensives	Defined according to JNC 7 (Chobanian et al. 2004)
	Normal BP (ODV in KHANES, NCV in NHANES)	SBP <120 mmHg and DBP <80 mmHg		SBP <120 mmHg and DBP <80 mmHg	
	Pre-HTN (ODV in KHANES, NCV in NHANES)	SBP ≥120 mmHg but <140 mmHg or DBP ≥80 mmHg but <90 mmHg		SBP ≥120 mmHg but <140 mmHg or DBP ≥80 mmHg but <90 mmHg	
	Anti-HTN medication (ODV)	Those participants currently taking anti-hypertensive medication	Questionnaire	Those participants now taking medication for high blood pressure	Questionnaire
	Mean SBP>140 mm Hg (NCV)	Mean SBP≥140 mmHg regardless of diagnosis of HTN or taking antihypertensives	Extracted from database	Mean SBP≥140 mmHg regardless of diagnosis of HTN or taking antihypertensives	Extracted from database
Laboratory analysis	**Glucose (mg/dL) (ODV)**	Glucose level in serum	Hitachi 7600 Automatic Analyser	Glucose level in serum	Hitachi Modular P Chemistry Analyzer
	Insulin (ulU/mL) (ODV)	Insulin level in serum	1470 WIZARD gamma counter immunoradiometric assay	Insulin level in serum	Human Insulin Immunoassay Using ROCHE ELECSYS 2010
	HbA1C(%) (ODV)	HbA1C level in serum	Tosoh G8 high-performance liquid chromatography	HbA1C level in serum	G7 Glycohemoglobin Analyzer
	Total cholesterol (mg/dL) (ODV)	Total cholesterol in serum	Hitachi 7600 Automatic Analyser	Total cholesterol in serum	Beckman Synchron LX20
	HDL (mg/dL) (ODV)	HDL in serum	Hitachi 7600 Automatic Analyser	HDL in serum	Roche/Hitachi Modular P Chemistry Analyzer
	LDL (mg/dL) (ODV)	LDL in serum	Hitachi 7600 Automatic Analyser	LDL in serum	Roche/Hitachi Modular P Chemistry

					Analyzer
	Triglycerides (mg/dL) (ODV)	Triglycerides in serum	Enzymatic method	Triglycerides in serum	Beckman UniCel® DxC800 Synchron
	hs-CRP (mg/dL) (ODV)	Hs-CRP in serum	Tosoh G8 high-performance liquid chromatography	hs-CRP in serum	Siemens/Behring Nephelometer II Analyzer System (BNII)
	WBC (thous/µL) (ODV)	WBC in serum	Laser flow cytometry using a Sysmex XE-2100D	WBC in serum	The Coulter HMX Hematology Analyzer
	Creatinine (mg/dL) (ODV)	Creatinine in serum	Hitachi 7600 Automatic Analyser	Creatinine in urine sample	Beckman UniCel® DxC800 Synchron
Periodontal measurement	**Periodontitis KNHANES-> (ODV)= CPI score; (NCV)= CPI cumulative and CPI continuous.** **NHANES-> (ODV=CDC/AAP definition, Severe/moderate/mild periodontitis)**	Community Periodontal Index **(CPI) ≥ 3 or 4 (high periodontal inflammation)** On six sites (mesio-buccal, mid-buccal, disto-buccal, mesio-lingual, mid-lingual and disto-lingual) per tooth using ten index teeth (#17, #16, #11, #26, #27, #46 and #47, #31, #36, #37). In absence of any index tooth, the adjacent remaining tooth in that sextant was examined and the highest score was recorded for that sextant	Trained dentists assessed the periodontal status using CPI probe with a 0.5-mm ball tip through a walking probe method. - CPI scores: 0 = normal periodontal tissue; 1 = gingival bleeding on probing; 2 = presence of calculus; 3 = probing pocket depth 4-5 mm; 4 = probing pocket depth ≥6 mm Inter-examiner reliability: Kappa value= 0.84 (Cheongwongun, 2016)	**-Severe periodontitis:** ≥ 2 interproximal sites with ≥ 6mm attachment loss (AL) (not on the same tooth) **AND** AL and 1 or more interproximal site with Probing depth (PD) ≥5 mm **-Moderate periodontitis:** ≥ 2 interproximal sites with ≥ 4mm AL (not on the same tooth) **OR** ≥ 2 interproximal sites with PD ≥5 mm (also not on the same tooth) **-Mild periodontitis:** ≥ 2 interproximal sites with ≥ 3mm AL **AND** or ≥ 2 more interproximal sites with ≥4 mm PD (not on the same tooth) or 1 site with ≥ 5 mm (Eke et al. 2012) **-Continuous measurements:** The extent of periodontal pockets (PPD) of ≥ 4mm, ≥5mm, ≥6 mm (number and percentage of sites) and b) extent of loss of periodontal attachment (AL) of ≥3mm, ≥ 4	-Trained dental hygienists carried out full mouth examination -Population: adults ≥30 yrs with 1 or more natural teeth. Third molars excluded -Bleeding or probing and furcations not registered Inter-examiner reliability: Kappa value= 0.70 and 0.71 for the two examiners (Eke et al. 2015)

				mm, ≥5mm, ≥6mm (number and percentage of sites).	
	CPI cumulative score	Sum of CPI scores of all sextants >2	This variables was created to assess the extent and severity of periodontitis	N/A	N/A
	CPI continuous score	Sum of CPI scores of all sextants			

ODV: Original database variable. As per definition in each specific database.
MDV: Modified database variable. Variable modified in order to group similar individuals in different databases.
NCV: Newly created variable based on information provided in each database.
NHANES, National Health and Nutrition Examination Survey; **KNHANES,** Korea National Health and Nutrition Examination Survey; **BP,** Blood Pressure; **SBP,** Systolic Blood Pressure; **DBP,** Diastolic Blood Pressure; **HTN,** Hypertension; **JNC 7,** Joint National Committee 7; **BMI,** Body Mass Index; **HbA1c,** Glycohemoglobin A1c; **HDL,** High-Density Lipoprotein; **LDL,** Low-Density Lipoprotein; **hs-CRP,** High Sensitivity C-reactive Protein; **WBC,** White Blood Cells.

Table S2 (Models A,B,C, D): Mediation analyses.

Model A (Unadjusted)	**Mediator 1: CRP**				**Mediator 2: WBC**				**Mediators: CRP +WBC**				
NHANES	**Effect Estimate**	**SE**	**95% CI**	**p-value**	**Effect Estimate**	**SE**	**95% CI**	**p-value**	**NHANES**	**Effect Estimate**	**SE**	**95% CI**	**p-value**
a (exposure → mediator)	0.189	0.043	(0.106, 0.273)	<0.001	0.342	0.096	(0.155, 0.529)	<0.001	a (exposure → mediator 1)	0.342	0.096	(0.155, 0.529)	<0.001
b (mediator → outcome)	0.053	0.006	(0.04, 0.065)	<0.001	0.004	0.003	(-0.001, 0.010)	0.193	b_1 (mediator 1 → mediator 2)	0.105	0.007	(0.090, 0.119)	<0.001
									b_2 (mediator 2 → outcome)	0.054	0.007	(0.041, 0.067)	<0.001
c (total effect)	0.160	0.016	(0.128, 0.192)	<0.001	0.160	0.016	(0.128, 0.192)	<0.001	c (total effect)	0.160	0.016	(0.128, 0.193)	<0.001
c' (direct effect)	0.150	0.016	(0.118, 0.182)	<0.001	0.158	0.016	(0.126, 0.191)	<0.001	c' (direct effect)	0.150	0.016	(0.118, 0.182)	<0.001
ab (mediated effect)	0.010	0.003	(0.005, 0.015)	<0.001	0.001	0.001	(-0.001, 0.003)	0.221	$ab_{1,2}$ (mediated effect)	0.002	0.001	(0.001, 0.003)	0.001
ab/c (percentage mediated) = %	0.067	0.019	(0.030, 0.104)	<0.001	0.008	0.007	(-0.005, 0.022)	0.228	ab1b2/c (percentage mediated) = %	0.013	0.004	(0.004, 0.021)	0.003
K-NHANES	**Effect Estimate**	**SE**	**95% CI**	**p-value**	**Effect Estimate**	**SE**	**95% CI**	**p-value**	**K-NHANES**	**Effect Estimate**	**SE**	**95% CI**	**p-value**
a (exposure → mediator)	0.224	0.076	(0.075, 0.372)	0.003	0.285	0.058	(0.170, 0.399)	<0.001	a (exposure → mediator 1)	0.285	0.058	(0.170, 0.339)	<0.001
b (mediator → outcome)	0.013	0.003	(0.007, 0.019)	<0.001	0.015	0.004	(0.007, 0.023)	<0.001	b_1 (mediator 1 → mediator 2)	0.237	0.020	(0.198, 0.276)	<0.001
									b_2 (mediator 2 → outcome)	0.012	0.003	(0.005, 0.018)	<0.001
c (total effect)	0.196	0.015	(0.166, 0.225)	<0.001	0.196	0.015	(0.166, 0.225)	<0.001	c (total effect)	0.192	0.015	(0.163, 0.222)	<0.001
c' (direct effect)	0.193	0.015	(0.163, 0.222)	<0.001	0.191	0.015	(0.162, 0.221)	<0.001	c' (direct effect)	0.190	0.015	(0.160, 0.219)	<0.001
ab (mediated effect)	0.003	0.001	(0.001, 0.005)	0.015	0.004	0.001	(0.001, 0.007)	0.004	$ab_{1,2}$ (mediated effect)	0.001	0.00	(0.000, 0.001)	0.004
ab/c (percentage mediated) = %	0.015	0.006	(0.003, 0.028)	0.017	0.022	0.008	(0.007, 0.037)	0.005	ab1b2/c (percentage mediated) = %	0.004	0.001	(0.001, 0.007)	0.006

NHANES: Exposure: CDC/AAP definition of periodontitis (categorical); Outcome: Hypertension (categorical)
KNHANES: Exposure: CPI-3/4 threshold for periodontitis (categorical); Outcome: Hypertension (categorical)

Model B (Unadjusted)	**Mediator 1: CRP**				**Mediator 2: WBC**				**Mediators: CRP + WBC**				
NHANES[1]	**Effect Estimate**	**SE**	**95% CI**	**p-value**	**Effect Estimate**	**SE**	**95% CI**	**p-value**	**NHANES[1]**	**Effect Estimate**	**SE**	**95% CI**	**p-value**
a (exposure → mediator)	0.166	0.034	(0.100, 0.232)	<0.001	0.426	0.075	(0.278, 0.573)	<0.001	a (exposure → mediator 1)	0.426	0.075	(0.0278, 0.573)	<0.001
b (mediator → outcome)	0.941	0.244	(0.463, 1.418)	<0.001	-0.003	0.109	(-0.218, 0.211)	0.975	b_1 (mediator 1 → mediator 2)	0.104	0.007	(0.089, 0.118)	<0.001
									b_2 (mediator 2 → outcome)	0.996	0.250	(0.505, 1.487)	<0.001
c (total effect)	4.600	0.482	(3.665, 5.545)	<0.001	4.600	0.482	(3.655, 5.545)	<0.001	c (total effect)	4.646	0.485	(3.696, 5.595)	<0.001
c' (direct effect)	4.444	0.483	(3.498, 5.390)	<0.001	4.602	0.484	(3.652, 5.551)	<0.001	c' (direct effect)	4.480	0.484	(3.531, 5.429)	<0.001
ab (mediated effect)	0.156	0.051	(0.056, 0.257)	0.002	-0.001	0.047	(-0.093, 0.090)	0.975	$ab_{1,2}$ (mediated effect)	0.044	0.014	(0.017, 0.071)	0.002
ab/c (percentage mediated) = %	0.035	0.012	(0.011, 0.060)	<0.001	0.000	0.01	(-0.020, 0.019)	0.975	$ab_{1,2}$/c (percentage mediated) = %	0.010	0.003	(0.003, 0.016)	0.003
NHANES[2]	**Effect Estimate**	**SE**	**95% CI**	**p-value**	**Effect Estimate**	**SE**	**95% CI**	**p-value**	**NHANES[2]**	**Effect Estimate**	**SE**	**95% CI**	**p-value**
a (exposure → mediator)	0.086	0.018	(0.052, 0.121)	<0.001	0.174	0.039	(0.097, 0.251)	<0.001	a (exposure → mediator 1)	0.174	0.039	(0.097, 0.251)	<0.001
b (mediator → outcome)	0.855	0.240	(0.384, 1.326)	<0.001	-0.016	0.107	(-0.227, 0.194)	0.878	b_1 (mediator 1 → mediator 2)	0.104	0.007	(0.090, 0.119)	<0.001
									b_2 (mediator 2 → outcome)	0.913	0.247	(0.429, 1.397)	<0.001
c (total effect)	3.478	0.249	(2.991, 3.966)	<0.001	3.478	0.249	(2.991, 3.966)	<0.001	c (total effect)	3.498	0.249	(3.01, 3.987)	<0.001
c' (direct effect)	3.405	0.249	(2.917, 3.893)	<0.001	3.481	0.249	(2.993, 3.970)	<0.001	c' (direct effect)	3.419	0.249	(2.930,3.908)	<0.001
ab (mediated effect)	0.074	0.026	(0.024, 0.124)	0.004	-0.003	0.019	(-0.040, 0.034)	0.878	$ab_{1,2}$ (mediated effect)	0.017	0.006	(.005, 0.028)	0.005
ab/c (percentage mediated) = %	0.022	0.008	(0.006, 0.037)	0.005	-0.001	0.005	(-0.011, 0.010)	0.878	$ab_{1,2}$/c (percentage mediated) = %	0.005	0.002	(0.001, 0.008)	0.007
K-NHANES	**Effect Estimate**	**SE**	**95% CI**	**p-value**	**Effect Estimate**	**SE**	**95% CI**	**p-value**	**K-NHANES**	**Effect Estimate**	**SE**	**95% CI**	**p-value**
a (exposure →	0.032	0.007	(0.017,	<0.001	0.042	0.006	(0.031,	<0.001	a (exposure →	0.042	0.006	(0.031,	<0.001

mediator)			0.046				0.053)		mediator 1)			0.053)	
b (mediator → outcome)	0.407	0.114	(0.182, 0.631)	<0.001	0.529	0.149	(0.238, 0.820)	<0.001	b_1 (mediator 1 → mediator 2)	0.233	0.020	(0.194, 0.273)	<0.001
									b_2 (mediator 2 → outcome)	0.344	0.116	(0.117, 0.572)	0.003
c (total effect)	0.611	0.053	(0.506, 0.715)	<0.001	0.611	0.053	(0.506, 0.715)	<0.001	c (total effect)	0.592	0.054	(0.486, 0.697)	<0.001
c' (direct effect)	0.598	0.053	(0.493, 0.702)	0.001	0.588	0.054	(0.483, 0.693)	<0.001	c' (direct effect)	0.581	0.054	(0.476, 0.686)	<0.001
ab (mediated effect)	0.013	0.005	(0.004, 0.022)	0.006	0.022	0.007	(0.009, 0.036)	0.001	$ab_{1,2}$ (mediated effect)	0.003	0.001	(0.001, 0.006)	0.007
ab/c (percentage mediated) = %	0.021	0.008	(0.006, 0.037)	0.008	0.038	0.013	(0.013, 0.063)	0.003	$ab_{1,2}$/c (percentage mediated) = %	0.006	0.002	(0.001, 0.010)	0.010

NHANES[1]: Exposure: mean probing pocket depth (mean PPD); Outcome: mean SBP
NHANES[2]: Exposure: mean clinical attachment level (mean CAL); Outcome: mean SBP
K-NHANES: Exposure: CPI-continuous; Outcome: mean SBP

Model C (Adjusted)	**Mediator 1: CRP**				**Mediator 2: WBC**				**Mediators: CRP +WBC**				
NHANES	**Effect Estimate**	**SE**	**95% CI**	**p-value**	**Effect Estimate**	**SE**	**95% CI**	**p-value**	**NHANES**	**Effect Estimate**	**SE**	**95% CI**	**p-value**
a (exposure → mediator)	0.189	0.043	(0.106, 0.273)	<0.001	0.342	0.096	(0.155, 0.529)	<0.001	a (exposure → mediator 1)	0.342	0.096	(0.155, 0.529)	<0.001
b (mediator → outcome)	0.039	0.006	(0.028, 0.050)	<0.001	0.008	0.003	(0.002, 0.013)	0.004	b_1 (mediator 1 → mediator 2)	0.105	0.007	(0.090, 0.119)	<0.001
									b_2 (mediator 2 → outcome)	0.037	0.006	(0.026, 0.049)	<0.001
c (total effect)	-0.091	0.021	(-0.131,-0.051)	<0.001	-0.103	0.021	(-0.144, -0.062)	<0.001	c (total effect)	-0.092	0.021	(-0.132, -0.052)	<0.001
c' (direct effect)	-0.098	0.021	(-0.139, -0.058)	<0.001	-0.106	0.021	(-0.147, -0.064)	<0.001	c' (direct effect)	-0.099	0.021	(-0.139, -0.059)	<0.001
ab (mediated effect)	0.007	0.002	(0.004, 0.011)	<0.001	0.003	0.001	(0.000, 0.005)	0.024	$ab_{1,2}$ (mediated effect)	0.001	0.000	(0.00, 0.002)	0.002
ab/c (percentage mediated) = %	-0.075	0.025	(-0.125, -0.026)	0.003	-0.024	0.012	(-0.047, -0.001)	0.037	ab/c (percentage mediated) = %	-0.013	0.005	(-0.024, -0.003)	0.009
K-NHANES	**Effect Estimate**	**SE**	**95% CI**	**p-value**	**Effect Estimate**	**SE**	**95% CI**	**p-value**	**K-NHANES**	**Effect Estimate**	**SE**	**95% CI**	**p-value**

a (exposure → mediator)	0.224	0.076	(0.075, 0.372)	0.003	0.285	0.058	(0.170, 0.399)	<0.001	a (exposure → mediator 1)	0.285	0.058	(0.170, 0.399)	<0.001
b (mediator → outcome)	0.008	0.003	(0.002, 0.014)	0.006	0.008	0.004	(0.000, 0.015)	0.050	b_1 (mediator 1 → mediator 2)	0.237	0.020	0.198, 0.276)	<0.001
									b_2 (mediator 2 → outcome)	0.008	0.003	(0.002, 0.014)	0.014
c (total effect)	0.155	0.015	(0.126, 0.184)	<0.001	0.155	0.015	(0.126, 0.184)	<0.001	c (total effect)	0.154	0.015	(0.125, 0.183)	<0.001
c' (direct effect)	0.154	0.015	(0.125, 0.183)	<0.001	0.153	0.015	(0.124, 0.182)	<0.001	c' (direct effect)	0.152	0.015	(0.123, 0.181)	<0.001
ab (mediated effect)	0.002	0.001	(0.000, 0.004)	0.044	0.002	0.001	(0.000, 0.005)	0.069	$ab_{1,2}$ (mediated effect)	0.001	0.000	(0.000, 0.001)	0.031
ab/c (percentage mediated) = %	0.012	0.006	(0.000, 0.024)	0.049	0.014	0.008	(-0.002, 0.030)	0.077	ab/c (percentage mediated) = %	0.003	0.002	(0.000, 0.006)	0.036

NHANES: Exposure: CDC/AAP definition of periodontitis (categorical); Outcome: Hypertension (categorical)
KNHANES: Exposure: CPI-3/4 threshold for periodontitis (categorical); Outcome: Hypertension (categorical)
Adjusted for (NHANES): Age, gender, ethnicity, smoking, education level, chronic medical condition; **(KNHANES):** Age, gender, BMI, education level, smoking, diabetes, alcohol consumption, creatinine, physical activity, chronic medical conditions

Model D (Adjusted)	**Mediator 1: CRP**				**Mediator 2: WBC**				**Mediators: CRP + WBC**				
NHANES[1]	**Effect Estimate**	**SE**	**95% CI**	**p-value**	**Effect Estimate**	**SE**	**95% CI**	**p-value**	**NHANES[1]**	**Effect Estimate**	**SE**	**95% CI**	**p-value**
a (exposure → mediator)	0.086	0.018	(0.052, 0.121)	<0.001	0.174	0.039	(0.097,0.251)	<0.001	a (exposure → mediator 1)	0.174	0.039	(0.097, 0.251)	<0.001
b (mediator → outcome)	0.733	0.225	(0.292, 1.175)	0.001	0.151	0.101	(-0.0406, 0.348)	0.134	b_1 (mediator 1 → mediator 2)	0.104	0.007	(0.090, 0.119)	<0.001
									b_2 (mediator 2 → outcome)	0.692	0.232	(0.238, 1.145)	<0.001
c (total effect)	2.168	0.289	(1.601, 2.735)	<0.001	2.068	0.290	(1.499, 2.637)	<0.001	c (total effect)	2.149	0.290	(1.581, 2.718)	<0.001
c' (direct effect)	2.105	0.290	(1.537, 2.673)	<0.001	2.042	0.291	(1.472, 2.612)	<0.001	c' (direct effect)	2.089	0.290	(1.521, 2.658)	<0.001
ab (mediated effect)	0.063	0.023	(0.018, 0.109)	0.007	0.026	0.019	(-0.010, 0.063)	0.155	$ab_{1,2}$ (mediated	0.013	0.005	(0.002, 0.023)	0.015

									effect)				
ab/c (percentage mediated) = %	0.030	0.012	(0.006, 0.054)	0.013	0.013	0.009	(-0.005, 0.031)	0.168	$ab_{1,2}$/c (percentage mediated) = %	0.006	0.003	(0.001, 0.011)	0.023
NHANES[2]	**Effect Estimate**	**SE**	**95% CI**	**p-value**	**Effect Estimate**	**SE**	**95% CI**	**p-value**	**NHANES[2]**	**Effect Estimate**	**SE**	**95% CI**	**p-value**
a (exposure → mediator)	0.166	0.034	(0.100, 0.232)	<0.001	0.426	0.075	(0.278, 0.573)	<0.001	a (exposure → mediator 1)	0.426	0.075	(0.278, 0.573)	<0.001
b (mediator → outcome)	0.706	0.225	(0.266, 1.147)	<0.001	0.138	0.101	(2.897, 4.700)	<0.001	b_1 (mediator 1 → mediator 2)	0.104	0.007	(0.089, 0.118)	<0.001
									b_2 (mediator 2 → outcome)	0.671	0.231	(0.505, 1.487)	<0.001
c (total effect)	3.894	0.459	(2.994, 4.739)	<0.001	3.857	0.458	(2.959, 4.766)	<0.001	c (total effect)	3.864	0.461	(3.696, 5.595)	<0.001
c' (direct effect)	3.776	0.460	(2.875, 4.678)	<0.001	3.798	0.460	(2.897, 4.700)	<0.001	c' (direct effect)	3.752	0.461	(3.531, 5.429)	<0.001
ab (mediated effect)	0.117	0.044	(0.031, 0.204)	0.008	0.059	0.044	(-0.028, 0145)	0.183	$ab_{1,2}$ (mediated effect)	0.030	0.012	(0.003, 0.016)	0.003
ab/c (percentage mediated) = %	0.031	0.013	(0.006, 0.056)	0.013	0.015	0.012	(-0.008, 0.039)	0.195	$ab_{1,2}$/c (percentage mediated) = %	0.008	0.003	(0.001, 0.014)	0.017
K-NHANES	**Effect Estimate**	**SE**	**95% CI**	**p-value**	**Effect Estimate**	**SE**	**95% CI**	**p-value**	**K-NHANES**	**Effect Estimate**	**SE**	**95% CI**	**p-value**
a (exposure → mediator)	0.032	0.007	(0.017, 0.046)	<0.001	0.042	0.006	(0.031, 0.053)	<0.001	a (exposure → mediator 1)	0.042	0.006	(0.031, 0.053)	<0.001
b (mediator → outcome)	0.237	0.111	(0.019, 0.455)	0.033	0.298	0.144	(0.015, 0.580)	0.039	b_1 (mediator 1 → mediator 2)	0.233	0.020	(0.194, 0.273)	<0.001
									b_2 (mediator 2 → outcome)	0.344	0.116	(0.117, 0.572)	0.003
c (total effect)	0.494	0.052	(0.391, 0.596)	<0.001	0.494	0.052	(0.391, 0.596)	<0.001	c (total effect)	0.591	0.054	(0.485, 0.696)	<0.001

c' (direct effect)	0.486	0.053	(0.383, 0.589)	<0.001	0.481	0.053	(0.378, 0.584)	<0.001	c' (direct effect)	0.581	0.054	(0.476, 0.686)	<0.001
ab (mediated effect)	0.007	0.004	(0.000, 0.015)	0.056	0.013	0.006	(0.000, 0.025)	0.047	$ab_{1,2}$ (mediated effect)	0.010	0.002	(0.007, 0.013)	<0.001
ab/c (percentage mediated) = %	0.015	0.008	(-0.001, 0.032)	0.064	0.026	0.014	(-0.001, 0.053)	0.058	$ab_{1,2}$/c (percentage mediated) = %	0.006	0.002	(0.001, 0.010)	0.010

NHANES[1]: Exposure: mean probing pocket depth (mean PPD); Outcome: mean SBP
NHANES[2]: Exposure: mean clinical attachment level (mean CAL); Outcome: mean SBP
K-NHANES: Exposure: CPI-continuous; Outcome: mean SBP
NHANES: Adjusted for: Age, gender, ethnicity, smoking, education level, chronic medical condition; **KNHANES: Adjusted for:** Age, gender, BMI, education level, smoking, diabetes, alcohol consumption, creatinine, physical activity, chronic medical conditions

The 3rd original publication

Hypertension

PERIODONTITIS

Association Between Periodontitis and Blood Pressure Highlighted in Systemically Healthy Individuals

Results From a Nested Case-Control Study

Eva Muñoz Aguilera, Jean Suvan, Marco Orlandi, Queralt Miró Catalina, Jose Nart, Francesco D'Aiuto

ABSTRACT: Recent evidence suggests hypertension and periodontitis are closely linked but limited data is available on the nature of the association. We aimed to investigate the relationship between periodontitis and mean arterial blood pressure in a sample of otherwise systemically healthy individuals. A case-control study including 250 cases (participants with periodontitis) and 250 controls (without periodontitis) was designed from a register of clinical trials conducted between 2000 and 2018 in a university setting. Cases were age, sex, and body mass index balanced with controls. Linear, logistic regression, and mediation models were planned to test the association between various periodontal measures and arterial blood pressure. We further investigated the role of systemic inflammation assessed by hs-CRP (high-sensitivity C-reactive protein) and white cell counts. Cases presented with 3.36 mm Hg (95% CI, 0.91–5.82, *P*=0.007) higher mean systolic blood pressure and 2.16 mm Hg (95% CI, 0.24–4.08, *P*=0.027) higher diastolic blood pressure than controls. Diagnosis of periodontitis was associated with mean systolic blood pressure (β=3.46±1.25, *P*=0.005) and greater odds of systolic blood pressure ≥140 mm Hg (odds ratio, 2.3 [95% CI, 1.15–4.60], *P*=0.018) independent of common cardiovascular risk factors. Similar findings were observed when continuous measures of periodontal status were modeled against systolic blood pressure. Measures of systemic inflammation although elevated in periodontitis were not found to be mediators of the association between periodontitis and arterial blood pressure values. Periodontitis is linked to higher systolic blood pressure in otherwise healthy individuals. Promotion of periodontal and systemic health strategies in the dental and medical setting could help reduce the burden of hypertension and its complications. **(*Hypertension*. 2021;77:1765–1774. DOI: 10.1161/HYPERTENSIONAHA.120.16790.)** • **Data Supplement**

Key Words: blood pressure ■ cardiovascular diseases ■ hypertension ■ inflammation ■ periodontitis

Elevated arterial blood pressure (BP) increases the risk of complications from cardiovascular diseases (CVD), such as stroke and myocardial infarction, with >7.6 million deaths accounted for every year and 143 million disability-adjusted life-years.[1] It is estimated that >30% of the overall population suffers from hypertension, and this estimate increases with age.[2] A 15% to 50% of individuals, however, are unaware they are affected by hypertension,[3] whereas many of those with a established diagnosis fail to achieve an optimal BP control despite their prescribed medications.[2] The burden and cost of hypertension remain high for any given society. Inflammation is considered an important driver of vascular dysfunction and implicated in the development and progression of hypertension.[4,5]

Periodontitis is a common inflammatory disease caused by a dysbiotic biofilm and affecting the soft and hard tissues around teeth.[6] It is a chronic disease, usually spanning over decades of an individual's life and is characterized by gingival inflammation with associated alveolar bone loss which, if not arrested, will ultimately lead to tooth loss. Almost 750 million people (aged 15–99 years) worldwide present with moderate to severe symptoms of periodontitis[7]; plus the disease

Correspondence to: Francesco D'Aiuto, Periodontology Unit, UCL Eastman Dental Institute, 21 University St, London, WC1E 6DE. Email f.daiuto@ucl.ac.uk
The Data Supplement is available with this article at https://www.ahajournals.org/doi/suppl/10.1161/HYPERTENSIONAHA.120.16790.
For Sources of Funding and Disclosures, see page 1773.

Hypertension is available at www.ahajournals.org/journal/hyp

Novelty and Significance

What Is New?

- The link between periodontitis and high blood pressure was confirmed in systemically healthy individuals.

What Is Relevant?

- Individuals with periodontitis but otherwise healthy presented with higher mean systolic blood pressure/diastolic blood pressure and odds of systolic blood pressure >140 mm Hg.
- Bleeding gums was associated with higher mean systolic blood pressure.
- Undetected hypertension was a common finding among the participants in this study.

Summary

The risk of elevated blood pressure was highlighted in systemically healthy patients with periodontitis. Oral health professionals could play a crucial role in assisting in the screening and management of hypertension.

Nonstandard Abbreviations and Acronyms

BMI	body max index
BP	blood pressure
CVD	cardiovascular diseases
DBP	diastolic BP
FMBS	full-mouth gingival bleeding
HDL	high-density lipoprotein
Hs-CRP	high-sensitivity C-reactive protein
IL	interleukin
LDL	low-density lipoprotein
PPD	probing pocket depths
SBP	systolic BP
TNF	tumor necrosis factor
WBC	white blood cell counts

is linked to social inequality and it negatively affects patients' quality of life.[8]

Recent evidence suggests a possible causal link between periodontitis and hypertension.[9] Patients with periodontitis often present with higher arterial BP values and a 30% to 70% higher chance to also present with hypertension,[10] especially when there is active gingival inflammation (ie, with gingival bleeding).[11] Longitudinal and large interventional studies confirming the nature of this association and the exact pathogenetic mechanisms are scarce.

The aim of this study was to investigate the association between diagnosis of severe periodontitis and arterial BP in a sample of otherwise healthy participants (without a confirmed diagnosis of hypertension). The primary objective was to assess office BP values in patients with periodontitis (cases) compared with controls (participants without periodontitis) and whether a linear relationship exists between measures of periodontitis extent/severity with BP values and whether basic measures of systemic inflammation mediate any association. Furthermore, the prevalence of undiagnosed hypertension between cases and controls was explored.

MATERIAL AND METHODS

The data that support the findings of this study are available from the corresponding author upon reasonable request.

Study Design and Population

A nested case-control study was designed, including participants recruited at the University College London Eastman Dental Institute between the years 2001 and 2018. All participants provided written informed consent at the time of study participation including use of data for future analyses. Seventeen clinical trials with the same inclusion criteria (severe periodontitis) or control (no periodontitis) and full-mouth periodontal assessment and arterial BP measurements (same recording protocol) were screened for inclusion in this analysis. This study was approved by the local UCL Research Ethics Committee (Project ID: 16989/001).

Sample Inclusion Criteria

Cases were individuals ≥18 years old who had been diagnosed with generalized severe periodontitis (defined as ≥50% of the teeth with probing pocket depths [PPD] of ≥5 mm and ≥30% marginal alveolar bone loss) and referred to the periodontal unit for management of their condition.[12] All participants were otherwise systemically healthy (no other systemic condition, as per medical history assessment/interview) and had not undertaken periodontal therapy within 6 months of the specific study assessment.

Controls were individuals ≥18 years old attending the same hospital without a diagnosis of periodontitis (recruited from other dental units but with no active dental infections) who were equally systemically healthy (no other systemic condition, as per medical history assessment/interview).

Sample Exclusion Criteria

Possible participants were excluded from this study if presenting with any of the following: (1) active infectious diseases, such as hepatitis, HIV, or tuberculosis, (2) any confirmed systemic diseases including diabetes, kidney, liver, cardiovascular diseases, hypertension, cancer, or on any chronic medication,

(3) pregnant or breastfeeding, and (4) taking nonsteroidal anti-inflammatory drugs on a regular basis or taking antibiotics within 3 months of assessment.

Periodontal Examination

Periodontal assessment used a standardized protocol carried out by calibrated examiners, as previously described.[12] Baseline data on full-mouth periodontal assessment was retrieved for all participants. Case definition of periodontitis was confirmed against the latest validated classification.[6] The full-mouth dental plaque and full-mouth gingival bleeding (FMBS) scores were recorded and the following thresholds for localized (FMBS 10%–29%) and generalized gingival bleeding (FMBS >30%) were adopted. Periodontitis case definition was of generalized severe/stage III/IV periodontitis.[6] Continuous measures of severity and extent of periodontitis were created as follows: (1) the extent of periodontal pockets with PPD of ≥4 mm, ≥5 mm, ≥6 mm (number and percentage of sites) and (2) extent of loss of periodontal attachment levels (clinical attachment level) of ≥3 mm, ≥4 mm, ≥5 mm, ≥6 mm (number and percentage of sites).

BP Assessment

Office BP measurements were obtained following a standardized protocol using an Omron device M5-1 (HEM-757A-E) by a trained person and recorded in triplicate for each participant, as previously described.[13] The patients were advised not to exercise, smoke, or consume any caffeine during the 30 minutes before their appointment. Upon arrival, the measurements were recorded after the patients were seated for 5 min and relaxed, with the back resting on the chair and the arm on a desk at the level of the right atrium. Average of the systolic and diastolic arterial pressure (systolic BP [SBP] and diastolic BP [DBP]) readings taken to the nearest value were obtained and used as continuous variables. Unconfirmed hypertension diagnosis was evaluated applying diagnostic thresholds of the US (values of SBP ≥130 mm Hg or DBP ≥80 mm Hg) and European (values of SBP ≥140 mm Hg or DBP ≥90 mm Hg) guidelines.[14,15]

Additional Variables

Socio-demographics information (age, sex, ethnicity), health lifestyle behavior (smoking, physical activity [frequency of weekly sessions of being active through walking, cycling, sports, and recreation]), and family history of cardiovascular diseases (whether or not any family member had heart or vascular disease) were retrieved from the medical history questionnaires. Anthropometric measurements (body max index: BMI) were collected by trained staff using a standard protocol.[12] Fasting venous blood samples were collected and analyzed for white blood cell (WBC) counts, hs-CRP (high-sensitivity C-reactive protein), total cholesterol, LDL and HDL (low- and high-density lipoprotein), glucose and triglycerides using standard biochemistry procedures and as previously described.[12]

Statistical Analysis

A sample size calculation confirmed that a minimum of 248 participants per group was required to detect a difference of 3.5 mm Hg in mean SBP between cases and controls, with an SD of 12 mm Hg, to achieve a power of at least 90% assuming an alpha of 0.05.

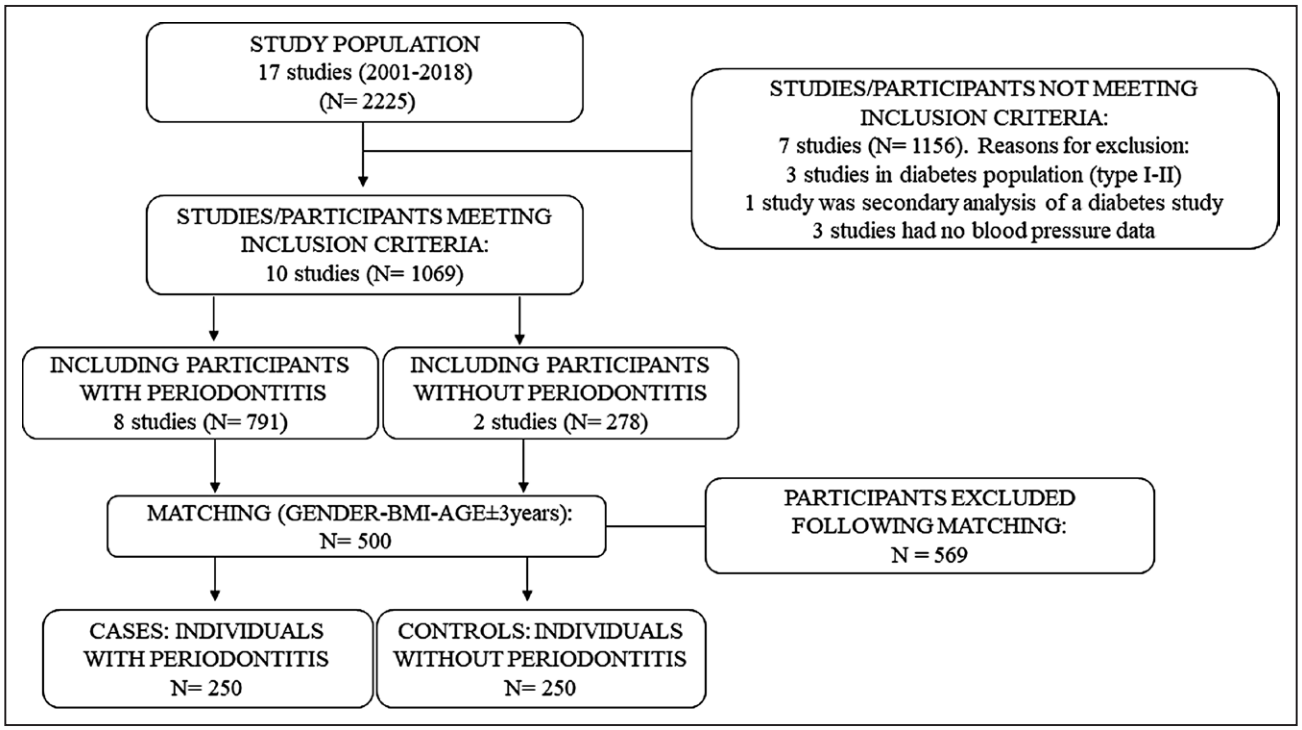


Figure. Study flow chart diagram.

Cases and controls were balanced based on age ±3years, sex, and BMI (Figure).[16] All data were checked for errors, entered in a single dataset, and analyzed using SPSS (version 25), STATA (version 15), and R Software (version 3.5.2). Descriptive statistics (comparisons between continuous variables used independent *t* test/Mann-Whitney and between categorical variables used χ^2 test) was used to compare general variables and arterial BP levels between cases and controls. Multivariate linear regression analysis was performed to investigate potential associations between (1) periodontitis (categorical and continuous definitions) or systemic markers of inflammation (ie, hs-CRP and WBC) as exposures and mean SBP and DBP as outcomes, (2) periodontitis (exposure) and systemic markers of inflammation (outcome). Age, BMI, sex, ethnicity, smoking, physical activity, and family history of CVD were used as confounders in each model. Similarly, multivariate logistic regression analysis was carried out to test the odds of the following categorical outcome variables: (1) SBP≥140, (2) SBP≥130, (3) DBP≥90, (4) DBP≥80 mm Hg, and undiagnosed hypertension defined as (5) per European guidelines[14] (SBP/DBP≥140/90 mmHg) and (6) as per US guidelines[15] (SBP/DBP≥130/80 mmHg) in relation to the different exposure variables, that is, periodontitis diagnosis (categorical yes/no), hs-CRP, and WBC. β coefficients and odds ratios with 95% CIs were calculated in unadjusted (model 1) and adjusted models including age, BMI, sex, ethnicity, smoking, physical activity, and family history of CVD (model 2). A *P* value of ≤0.05 was considered statistically significant. Mediation analyses of systemic inflammatory markers were performed if a positive association between CRP or WBC variables with periodontitis and SBP/DBP were found in linear/logistic regression models. Two different and prespecified routes were used; direct (route 1) and indirect (route 2) mediation effects with their 95% CI were estimated: route 1: periodontitis (exposure)→hypertension (outcome). Route 2: periodontitis (exposure)→WBC or hs-CRP (mediators)→hypertension (outcome).

RESULTS

Sample median age [interquartile range] was 35 [12] years with no substantial differences observed between cases (35 [9] years) and controls (34 [14] years; *P*=0.345). Similar sex (*P*=0.929) and BMI (*P*=0.209) distributions were confirmed between cases and controls. Higher number of current smokers, with predominant Black/Afro-Caribbean participants, reporting less physical activity and a higher percentage of family history of CVD were observed in cases when compared with controls. Participants with periodontitis exhibited increased glucose, LDL, CRP, and WBC levels but lower HDL values when compared with controls (all, *P*<0.01; Table 1).

Higher mean differences in SBP (3.36 mmHg [95% CI, 0.91–5.82], *P*=0.007) and DBP (2.02 mm Hg [95% CI, 0.24–4.08], *P*<0.027) were observed in cases when compared with controls.

A 14% of cases presented with SBP≥140 mm Hg versus a 7% of the controls (*P*=0.021). Similarly, >43% of the cases presented with DBP≥80 mm Hg versus 34% of the controls (*P*=0.035). The percentages of cases with SBP≥130 mm Hg or DBP≥90 mm Hg were greater than for the controls but not statistically significant (Table 1). Overall a 15.6% of the whole study participants presented with values of SBP/DBP in the range of hypertension (European definition): 17.2% of the cases and 14% of the controls (*P*=0.324) and a 45.6% (American definition): almost 50% of cases and a 41.6% of controls (*P*=0.073; Table 1).

Linear regression analysis confirmed an association between case definition of periodontitis (categorical) and higher mean SBP after adjusting for common risk factors (β±SE=3.46±1.25, *P*=0.005, model 2) and to DBP (β±SE=2.16±0.98, *P*=0.027, model 1; Table 2). Greater severity of periodontitis as assessed by mean clinical attachment level, PPD, and FMBS were associated with higher mean SBP (models 1 and 2) and with higher mean DBP (model 1; Table 2). A similar linear association between different thresholds of periodontal lesions (PPD thresholds) and SBP (models 1 and 2) were noted but for different threshold of clinical attachment level, this was only seen in model 1 (Table 2). Periodontitis (categorical and continuous variables) was associated with higher CRP (model 1) and WBC (model 1 and 2) levels (Table 2). Similarly, periodontitis (categorical) was associated with higher odds of SBP≥140 mm Hg (odds ratio, 1.98 [95% CI, 1.10–3.65], *P*=0.023, model 1; odds ratio, 2.31 [95% CI, 1.17–4.67], *P*=0.018, model 2) and DBP≥80 mm Hg (odds ratio, 1.47 [95% CI, 1.03–2.12], *P*=0.035, model 1; Table 3). No significant association was found with SBP≥130 mm Hg, DBP≥90 mm Hg, or Hypertension definition according to European or American guidelines.

When CRP and WBC were modeled as exposure variables, no associations were observed with SBP or DBP (Tables 2 and 3). Lastly, an association between SBP and increasing FMBS irrespective of periodontal diagnosis was observed in the multivariate fitted model for SBP according to bleeding status (Table 4). Participants with generalized bleeding presented with a 5 mm Hg greater SBP than those with healthy gums (95%CI, 8.22–1.91, *P*=0.002; Table 4).

Mediation analyses confirmed that WBC did not act as a mediator of the association between periodontitis (categorical) and SBP (continuous) in either in the unadjusted (β±SE=−0.00±1.21; *P*=0.994) or the fully adjusted (*β*±SE=−0.03±0.21; *P*=0.900) models (route 2; Tables S1 and S2 in the Data Supplement). Similar results were observed when the model was replicated for continuous periodontal (FMBS, clinical attachment level, PPD) and categorical BP (SBP≥140 mm Hg) variables.

DISCUSSION

The results of this study showed that systemically healthy individuals with periodontitis (cases) presented with higher mean SBP and DBP than participants without

Table 1. Main Characteristics of Participants

Variables	Overall (500)	Controls (Non-periodontitis; 250)	Cases (Periodontitis; 250)	*P* value
Categorical N (%)				
Sex (%female)*	263 (52.6)	132 (52.8)	131 (52.4)	0.929
Smoking				
Non-smoker	290 (58.0)	175 (70.0)	115 (46.0)	<0.001
Current smoker	96 (19.2)	34 (13.6)	62 (24.8)	<0.001
Ex-smoker	114 (22.8)	41 (16.4)	73 (29.2)	<0.001
Ethnicity				
White	299 (59.8)	177 (70.8)	122 (40.8)	<0.001
Asian	94 (18.8)	44 (17.6)	50 (20.0)	<0.001
Black-African	47 (9.4)	16 (6.4)	31 (12.4)	<0.001
Black-Caribbean	41 (8.2)	8 (3.2)	33 (13.2)	<0.001
Other	19 (3.8)	5 (2.0)	14 (5.6)	<0.001
Physical activity				
Daily	62 (12.4)	39 (15.6)	23 (9.2)	<0.001
> twice a week	178 (35.6)	118 (47.2)	60 (24.0)	<0.001
Once a week	63 (12.6)	30 (12.0)	33 (13.2)	<0.001
< once a week	27 (5.4)	4 (1.6)	23 (9.2)	<0.001
Never/rarely	170 (34.0)	59 (23.6)	111 (44.4)	<0.001
Family history of CVD	156 (31.2)	67 (26.8)	89 (35.6)	0.034
SBP≥140 mm Hg	54 (10.8)	19 (7.6)	35 (14.0)	0.021
DBP≥90 mm Hg	57 (11.4)	28 (11.2)	29 (11.6)	0.888
SBP≥130 mm Hg	138 (27.6)	65 (47.1)	73 (52.9)	0.424
DBP≥80 mm Hg	195 (39.0)	86 (34.4)	109 (43.6)	0.035
Hypertension definition (European guidelines)	78 (15.6)	35 (14.0)	43 (17.2)	0.324
Hypertension definition (American guidelines)	228 (45.6)	104 (41.6)	124 (49.6)	0.073
Localized gingivitis (FMBS 10%–29%)	176 (35.2)	143 (57.2)	33 (13.2)	<0.001
Generalized gingivitis (FMBS≥30)	252 (50.4)	40 (16.0)	212 (84.8)	<0.001
Continuous: mean (SD) or median [IQR]				
Age, y*	35 [12]	34 [14]	35 [9]	0.345
BMI, kg/m²*	24.01 [5.11]	23.75 [4.92]	24.42 [5.18]	0.209
SBP, mm Hg	122.39 (14.05)	120.70 (12.59)	124.07 (14.33)	0.007
DBP, mm Hg	77.02 (10.9)	75.94 (10.76)	78.10 (11.10)	0.027
Glucose, mmol/L	1.4 [2.09]	1.2 [1.1]	1.6 [3.30]	<0.001
Total cholesterol, mmol/L	5.05 [1.30]	5.01 [1.30]	5.10 [1.21]	0.091
HDL, mmol/L	1.56 (0.43)	1.64 (0.42)	1.49 (0.44)	<0.001
LDL, mmol/L	2.96 (0.83)	2.83 (0.84)	3.09 (0.79)	<0.001
Triglycerides, mmol/L	1.3 [1.49]	1.2 [1.10]	1.3 [2.10]	0.133
Hs-CRP, mg/L	1.7 [2.39]	1.38 [2.47]	1.86 [2.37]	0.008
WBC, 1000/μL	5.93 [2.20]	5.58 [1.99]	6.36 (2.26)	<0.001
PPD	2.56 [1.95]	1.99 [0.43]	3.94 [1.24]	<0.001
CAL	2.74 [2.24]	2.02 [0.47]	4.26 [1.52]	<0.001
FMBS	30.65 [26.74]	15.45 [15.27]	53.66 (33.37]	<0.001
FMPS	50.28 [39.38]	49.15 [40.78]	51.11 [39.43]	0.166

BMI indicates body mass index; CAL, clinical attachment level; CVD, cardiovascular diseases; DBP, diastolic blood pressure; FMBS, full-mouth gingival bleeding score; FMPS, full-mouth dental plaque score; HDL, high-density lipoprotein; Hs-CRP, high-sensitive C-reactive protein; IQR, Interquartile range; LDL, low-density lipoprotein; PPD, probing pocket depth; SBP, systolic blood pressure; and WBC, white blood cells.

*Cases and controls groups balanced according to age, sex, and BMI.

Table 2. Linear Regression Models of SBP, DBP and hs-CRP, WBC According to Various Indices of Periodontitis or Systemic Inflammation

Linear regression models				
	SBP		DBP	
Exposure: periodontitis	Model 1 β±SE	Model 2 β±SE	Model 1 β±SE	Model 2 β±SE
Categorical variable				
Cases vs controls (perio vs non perio)	3.36±1.25*	3.46±1.25*	2.16±0.98†	1.48±1.04
Continuous variables				
CAL (mean)	1.51±0.42‡	0.89±0.42†	0.69±0.33†	0.14±0.35
No. of sites with CAL ≥3 mm	0.03±0.01*	0.02±0.01	0.02±0.01	0.0002±0.01
No. of sites with CAL ≥4 mm	0.04±0.01*	0.02±0.01	0.02±0.01	0.004±0.01
No. of sites with CAL ≥5 mm	0.04±0.01*	0.02±0.01	0.02±0.01	0.006±0.01
No. of sites with CAL ≥6 mm	0.06±0.02*	0.03±0.02	0.03±0.02	0.004±0.01
% of sites with CAL ≥3 mm	0.06±0.02*	0.03±0.02	0.02±0.02	-0.006±0.01
% of sites with CAL ≥4 mm	0.06±0.02*	0.03±0.02	0.03±0.02	0.003±0.02
% of sites with CAL ≥5 mm	0.07±0.02*	0.04±0.02	0.03±0.02	0.01±0.02
% of sites with CAL ≥6 mm	0.10±0.03*	0.05±0.03	0.04±0.03	0.01±0.02
PPD (mean)	1.86±0.52‡	1.51±0.5*	0.81±0.41†	0.32±0.44
No. of sites with PPD ≥4 mm	0.04±0.01*	0.03±0.01†	0.02±0.01	0.008±0.01
No. of sites with PPD ≥5 mm	0.05±0.02*	0.04±0.02†	0.02±0.01	0.01±0.01
No. of sites with PPD ≥6 mm	0.08±0.02*	0.06±0.02*	0.03±0.02	0.01±0.02
% of sites with PPD ≥4 mm	0.07±0.02*	0.05±0.02†	0.03±0.02	0.01±0.02
% of sites with PPD ≥5 mm	0.07±0.03*	0.06±0.03†	0.03±0.02	0.01±0.02
% of sites with PPD ≥6 mm	0.12±0.04*	0.10±0.04*	0.05±0.03	0.02±0.03
FMBS (mean)	0.09±0.02‡	0.07±0.02*	0.04±0.02†	0.02±0.02
Exposure: systemic inflammation	SBP		DBP	
	Model 1 β±SE	Model 2 β±SE	Model 1 β±SE	Model 2 β±SE
Hs-CRP	0.22±0.19	0.18±0.17	0.24±0.15	0.21±0.14
WBC	0.17±0.38	0.61±0.34	0.25±0.30	0.44±0.28
Exposure: periodontitis	Hs-CRP (log hs-CRP)		WBC (log WBC)	
	Model 1 β±SE	Model 2 β±SE	Model 1 β±SE	Model 2 β±SE
Categorical variable				
Cases vs controls (perio vs nonperio)	0.24±0.08*	0.01±0.09	0.08±0.23‡	0.08±0.02*
Continuous variables				
CAL (mean)	0.083±0.029*	0.040±0.032	0.022±0.008*	0.023±0.008*
PPD (mean)	0.112±0.036*	0.05±0.04	0.031±0.01*	0.029±0.01*
FMBS (mean)	0.004±0.002†	0.002±0.002	0.002±0.000‡	0.001±0.000*

Model 1: Unadjusted. Model 2: age, BMI, sex, ethnicity, smoking, physical activity, family history of CVD. B indicates beta coefficient; CAL, clinical attachment level; DBP, diastolic blood pressure; FMBS, full-mouth gingival bleeding score; Hs-CRP, high-sensitive C-reactive protein; PPD, probing pocket depth; SBP, systolic blood pressure; and WBC, white blood cells.

*$P<0.01$.

†$P<0.05$.

‡$P<0.001$.

periodontitis (controls). Cases presented with more than twice higher likelihood of SBP≥140 mm Hg and almost 50% higher odds of DBP≥80 mm Hg than the controls.

A recent systematic review confirmed a 4.5 mm Hg SBP and 2.5 mm Hg DBP greater arterial BP values in the general population (including participants with and without other comorbidities).[10] In a previously reported age- and sex-matched case-control study looking at the association of periodontitis, systemic inflammation, and endothelial function, greater differences in SBP (7 mm Hg) and CRP (1.3 mg/L) levels between the cases and controls were observed.[17]

Elevated BP remains the main risk factor for heart failure, atrial fibrillation, chronic kidney disease, heart valve diseases, aortic syndromes, and dementia, in addition to coronary heart disease and stroke.[18] It is now

Table 3. Multiple Logistic Regression Models of SBP≥140 mm Hg, DBP≥90 mm Hg, SBP≥130 mm Hg, DBP≥80 mm Hg and Hypertension Definitions according to Periodontitis Diagnosis, hs-CRP and WBC

Multiple logistic models			
Exposure: periodontitis	SBP≥140 mm Hg	DBP≥90 mm Hg	Hypertension definition (European)
Model 1 OR (95% CI)	1.98 (1.10–3.65)*	1.05 (0.59–1.81)	1.28 (0.78–2.08)
Model 2 OR (95% CI)	2.31 (1.17–4.67)*	1.05 (0.55–1.98)	1.24 (0.71–2.18)
Exposure: Hs-CRP			
Model 1 OR (95% CI)	0.98 (0.89–1.08)	0.97 (0.88–1.08)	0.97 (0.88–1.05)
Model 2 OR (95% CI)	0.95 (0.84–1.06)	0.97 (0.86–1.09)	0.95 (0.85–1.05)
Exposure: WBC			
Model 1 OR (95% CI)	1.00 (0.84–1.18)	0.92 (0.78–1.10)	0.97 (0.84–1.13)
Model 2 OR (95% CI)	1.05 (0.87–1.27)	0.97 (0.80–1.17)	1.02 (0.87–1.20)
Exposure: periodontitis	SBP≥130 mm Hg	DBP≥80 mm Hg	Hypertension definition (American)
Model 1 OR (95% CI)	1.17 (1.17–1.73)	1.47 (1.03–2.12)*	1.38 (0.97–1.96)
Model 2 OR (95% CI)	1.26 (0.78–2.00)	1.20 (0.78–1.84)	1.23 (0.81–1.88)
Exposure: Hs-CRP			
Model 1 OR (95% CI)	1.02 (0.96–1.08)	1.06 (0.99–1.12)	1.06 (0.99–1.13)
Model 2 OR (95% CI)	1.02 (0.96–1.08)	1.06 (0.99–1.12)	1.06 (0.99–1.27)
Exposure: WBC			
Model 1 OR (95% CI)	1.00 (0.89–1.13)	1.04 (0.93–1.16)	1.03 (0.93–1.15)
Model 2 OR (95% CI)	1.06 (0.93–1.21)	1.08 (0.96–1.22)	1.09 (0.97–1.23)

Model 1: unadjusted. Model 2: age, BMI, sex, ethnicity, smoking, physical activity, family history of CVD. BMI indicates body mass index; CVD, cardiovascular diseases; DBP, diastolic blood pressure; Hs-CRP, high-sensitive C-reactive protein; OR, odds ratio; SBP, systolic blood pressure; and WBC, white blood cells.

*$P<0.01$.

†$P<0.05$.

‡$P<0.001$.

understood that biologically normal BP levels are lower than what previously fell within a normal range.[15] The observed differences of SBP/DBP between cases and control in this study could be clinically relevant and might represent an overlooked mechanism linking periodontitis with increased future CVD risk.[19]

In agreement with previous studies,[20,21] this study showed that irrespective of periodontal status, bleeding gums was associated with SBP. Similarly, a more recent secondary analysis of NHANES (III also reported a 2.6 mm Hg higher mean SBP for gingivitis, also independently associated with 40% greater odds of high/uncontrolled BP.[11] Bleeding gums, the earliest sign of periodontal diseases, has also been linked to increased systemic biomarkers and vascular changes.[22,23] Self-report of bleeding gums is an easy measure of periodontal inflammation for both patients and clinicians and it could represent a valuable parameter in routine BP screening protocols.

Several mechanisms underlying the links between gingival diseases and hypertension have been proposed with dysbiotic subgingival microbiome triggering low-grade systemic inflammation and oxidative stress representing the main pathways.[24] Periodontitis patients express not only increased local and systemic inflammatory markers, such as CRP, TNF (tumor necrosis

Table 4. Multivariate Fitted SBP According to Bleeding Status

Bleeding status (irrespective of periodontitis)	N (%)	SBP Mean (SD)	Multiple comparisons ΔSBP (95% CI) *P* value		
			Gingival health	Localized bleeding	Generalized bleeding
Gingival health (FMBS<10%)	72 (14.4)	118.71 (11.23)	…		
Localized bleeding (10%>FMBS≤30%)	176 (35.2)	121.92 (14.68)	3.21 (6.61–0.20) *P*=0.065	…	
Generalized bleeding (FMBS≥30%)	252 (50.4)	123.77 (14.16)	5.06 (8.22–1.91) *P*=0.002	1.85 (4.65–0.94) *P*=0.192	…

ΔSBP indicates mean difference in SBP; FMBS, Full-mouth gingival bleeding score; and SBP, systolic blood pressure.

factor)-α, neutrophilic enzymes, WBC, and disparity in T-cell subtypes, but also neutrophil dysfunction, which are all mechanisms resulting in vascular changes and endothelial dysfunction.[19,25,26] The presence of periodontal pathogens has been linked to hypertension in epidemiological studies.[27] Preclinical evidence originated by experimental animal models, including immunizations with *Porphyromonas gingivalis* lysate and lipopolysaccharide-endotoxin from other gram-negative bacteria, caused prolonged T-cell activation and elicited increased levels of CRP, TNF-α, and IL (interleukin)-1β, resulting in increased BP.[28] Interaction between oral-gut microbiome can also contribute to amplification of inflammation and metabolic changes.[29] Recent evidence implicates oral bacteria in the nitrate-nitrite-nitric oxide (NO) pathway and pathogenesis of hypertension,[30] with high concentrations of nitrite-reductase bacteria increasing systemic NO and having an effect of lowering SBP.[31]

In the current study, hs-CRP/WBC as a proxy of systemic inflammation was associated with periodontitis but not with SBP/DBP. Additionally, WBC did not show a mediation effect between periodontitis and BP. These results are in partial disagreement with a recent analysis of cross-sectional data, based on national health surveys in US and Korea, where a 2% to 7% mediating effect of WBC and CRP was observed when examining the association between periodontitis and hypertension.[32] A possible explanation for these differences relates to an overall younger population of this study sample (35 years old) versus 51 and 46 years old in the American and Korean populations, and possibly due to the systemically healthy status of this sample, when compared with representative samples of those populations, including systemic conditions. Nevertheless, an association of arterial BP with both continuous and categorical measures of periodontitis in younger and systemically healthy individuals strengthens the evidence in favor of a causal association between the two diseases.[24]

A recent Mendelian Randomization analysis and results from a short-term pilot randomized controlled clinical trial on periodontal treatment of resistant hypertensive patients corroborate these findings.[9] Single nucleotide polymorphisms in genes *SIGLEC5, DEFA1A3, MTND1P5,* and *LOC107984137* loci in Genome-wide association studies (GWAS) linked to periodontitis, and BP phenotype were discovered and a noticeable reduction in SBP/DBP, endothelial function as well as inflammatory cytokines and activated T-cell subsets was observed 2 months following the treatment. Similarly, another randomized controlled clinical trial with 6 months follow-up on a prehypertensive population also observed a significant reduction in SBP/DBP following nonsurgical periodontal treatment.[33] Oral health promotion strategies such as tooth brushing twice daily has demonstrated very effective not only in managing and preventing most common oral conditions[34] but also in providing a powerful and affordable tool for hypertension control.[35] Notably, a 14% reduction in cardiovascular events has been observed with a 4.4 mm Hg reduction in SBP.[36] Preliminary evidence suggests that periodontal treatment in patients with type 2 diabetes, a common comorbidity, could result in substantial long-term reduction of medical-related costs for healthcare systems.[37] Thus, given the importance of nonpharmacological and pharmacological BP-lowering strategies in decreasing CVD risk and mortality,[38] larger multicenter randomized controlled clinical trials and health-economic analyses are warranted to further investigate the benefits of periodontal treatments on BP prevention and control.

Elevated BP is usually asymptomatic and best detected in screening programs or opportunistic measurements of BP, which confirm that a worryingly high number of individuals (>50%) is unaware of a possible diagnosis of hypertension.[39] The presented study confirmed that a 15% to 45% of the sample could exhibit undetected hypertension (depending on whether a European or US guideline definition was used), with 54% to 55% of these having periodontitis. In a recent cross-sectional study on the association of periodontitis and hypertension, a 15.9% of the study sample presented with undiagnosed high BP (based on a single office measurement), of which a 62.5% had periodontitis.[40] These data confirm that programs of hypertension screening in the dental settings should not be underestimated.

Although this study improves the understanding of the association between periodontitis and arterial BP, it is recognized some limitations exist. The study design and analysis may have introduced some bias namely through selection and assessment biases.[41] Furthermore, in this study we did not account for other factors that might have impacted BP, such as abdominal obesity, salt intake, use of anti-inflammatory drugs, hormone treatments, or stress as well as additional oral diseases (ie, caries). Future analyses should focus on existing or new epidemiological evidence (longitudinal studies) where all possible confounders are appropriately considered. Nevertheless, this study benefits from a robust research methodology in assessing the exposure (periodontitis) and outcome (BP), and sufficient statistical power could have counteracted some of the limitations.[42] Furthermore, using a balanced study design through matching for common confounders of arterial BP facilitated analysis of comparable groups.[43]

Conclusions

This study expands current knowledge on the association between periodontitis and elevated BP, pointing at the importance of this link in the generally healthy population. Oral health professionals could play a pivotal role in helping the medical community detecting and tackling the burden and consequences of hypertension.

Perspectives

Periodontal treatments could be well-tolerated novel nonpharmacological interventions for the management of hypertension. Particularly so when patients are informed that periodontal treatment could be beneficial not only for their oral health but also for their general health and wellbeing in return. Thus, future directions and broad implications of this work will involve liaison of dental and medical health professionals with the following objectives:

1. Raising awareness of the increased risk for high BP among individuals with periodontal diseases.
2. Implementing hypertension screening systems by dental professionals and prompt referral to general practitioners.
3. Implementing periodontal disease screening systems by medical professionals and referral to dental practitioners.
4. Providing advice for common risk factors: Healthy diet, smoking cessation, promoting physical activity, alcohol reduction, and diabetes management.
5. Early diagnosis and management of gingivitis and periodontitis. Effective prevention and treatment of these conditions is very cost-effective and has shown an effect in reduction of systemic markers of inflammation and improvement in endothelial function.
6. Future research will involve larger multicenter randomized controlled clinical trials to test the effects on periodontal treatment on BP levels.

ARTICLE INFORMATION

Received December 14, 2020; accepted February 2, 2021.

Affiliations

From the Periodontology Unit, UCL Eastman Dental Institute and Hospital, University College London, United Kingdom (E.M.A., J.S., M.O., F.D.); and Department of Periodontology, Universitat Internacional de Catalunya, Barcelona, Spain (E.M.A., Q.M.C., J.N.).

Acknowledgments

We acknowledge that contribution of this work received a proportion of funding from the UK Department of Health's National Institute for Health Research Biomedical Research Centre at University College London/University College London Hospitals. The Graphic abstract (online-only) was created with BioRender.com.

Sources of Funding

M. Orlandi holds a National Institute for Health Research (NIHR) Clinical Lectureship. F. D'Aiuto held a Clinical Senior Lectureship Award supported by the UK Clinical Research Collaboration.

Disclosures

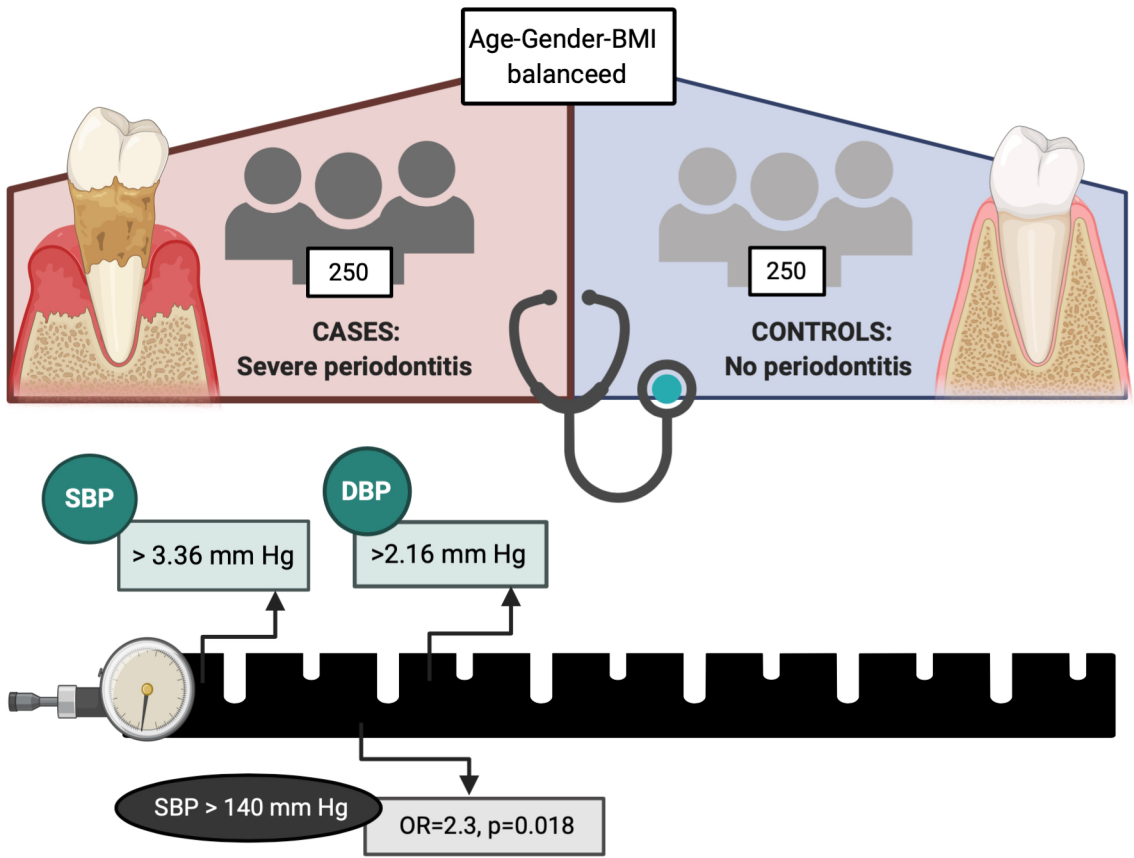
None.

REFERENCES

1. Forouzanfar MH, Afshin A, Alexander LT; GBD 2015 Risk Factors Collaborators. Global, regional, and national comparative risk assessment of 79 behavioural, environmental and occupational, and metabolic risks or clusters of risks, 1990–2015: a systematic analysis for the Global Burden of Disease Study 2015. *Lancet.* 2016;388:1659–1724. doi: 10.1016/S0140-6736(16)31679-8
2. Mills KT, Bundy JD, Kelly TN, Reed JE, Kearney PM, Reynolds K, Chen J, He J. Global disparities of hypertension prevalence and control: a systematic analysis of population-based studies from 90 Countries. *Circulation.* 2016;134:441–450. doi: 10.1161/CIRCULATIONAHA.115.018912
3. Scholes S, Conolly A, Mindell JS. Income-based inequalities in hypertension and in undiagnosed hypertension: analysis of Health Survey for England data. *J Hypertens.* 2020;38:912–924. doi: 10.1097/HJH.0000000000002350
4. Drummond GR, Vinh A, Guzik TJ, Sobey CG. Immune mechanisms of hypertension. *Nat Rev Immunol.* 2019;19:517–532. doi: 10.1038/s41577-019-0160-5
5. Jayedi A, Rahimi K, Bautista LE, Nazarzadeh M, Zargar MS, Shab-Bidar S. Inflammation markers and risk of developing hypertension: a meta-analysis of cohort studies. *Heart.* 2019;105:686–692. doi: 10.1136/heartjnl-2018-314216
6. Tonetti MS, Greenwell H, Kornman KS. Staging and grading of periodontitis: framework and proposal of a new classification and case definition. *J Periodontol.* 2018;89(suppl 1):S159–S172. doi: 10.1002/JPER.18-0006
7. Frencken JE, Sharma P, Stenhouse L, Green D, Laverty D, Dietrich T. Global epidemiology of dental caries and severe periodontitis - a comprehensive review. *J Clin Periodontol.* 2017;44(suppl 18):S94–S105. doi: 10.1111/jcpe.12677
8. Ferreira MC, Dias-Pereira AC, Branco-de-Almeida LS, Martins CC, Paiva SM. Impact of periodontal disease on quality of life: a systematic review. *J Periodontal Res.* 2017;52:651–665. doi: 10.1111/jre.12436
9. Czesnikiewicz-Guzik M, Osmenda G, Siedlinski M, Nosalski R, Pelka P, Nowakowski D, Wilk G, Mikolajczyk TP, Schramm-Luc A, Furtak A, et al. Causal association between periodontitis and hypertension: evidence from Mendelian randomization and a randomized controlled trial of non-surgical periodontal therapy. *Eur Heart J.* 2019;40:3459–3470. doi: 10.1093/eurheartj/ehz646
10. Muñoz Aguilera E, Suvan J, Buti J, Czesnikiewicz-Guzik M, Barbosa Ribeiro A, Orlandi M, Guzik TJ, Hingorani AD, Nart J, D'Aiuto F. Periodontitis is associated with hypertension: a systematic review and meta-analysis. *Cardiovasc Res.* 2020;116:28–39. doi: 10.1093/cvr/cvz201
11. Pietropaoli D, Monaco A, D'Aiuto F, Muñoz Aguilera E, Ortu E, Giannoni M, Czesnikiewicz-Guzik M, Guzik TJ, Ferri C, Del Pinto R. Active gingival inflammation is linked to hypertension. *J Hypertens.* 2020;38:2018–2027. doi: 10.1097/HJH.0000000000002514
12. D'Aiuto F, Parkar M, Nibali L, Suvan J, Lessem J, Tonetti MS. Periodontal infections cause changes in traditional and novel cardiovascular risk factors: results from a randomized controlled clinical trial. *Am Heart J.* 2006;151:977–984. doi: 10.1016/j.ahj.2005.06.018
13. Muntner P, Shimbo D, Carey RM, Charleston JB, Gaillard T, Misra S, Myers MG, Ogedegbe G, Schwartz JE, Townsend RR, et al. Measurement of blood pressure in humans: a scientific statement from the American Heart Association. *Hypertension.* 2019;73:e35–e66. doi: 10.1161/HYP.0000000000000087
14. Williams B, Mancia G, Spiering W, Agabiti Rosei E, Azizi M, Burnier M, Clement DL, Coca A, de Simone G, Dominiczak A, et al; ESC Scientific Document Group. 2018 ESC/ESH Guidelines for the management of arterial hypertension. *Eur Heart J.* 2018;39:3021–3104. doi: 10.1093/eurheartj/ehy339
15. Whelton PK, Carey RM, Aronow WS, Casey DE Jr, Collins KJ, Dennison Himmelfarb C, DePalma SM, Gidding S, Jamerson KA, Jones DW, et al. 2017 ACC/AHA/AAPA/ABC/ACPM/AGS/APhA/ASH/ASPC/NMA/PCNA guideline for the prevention, detection, evaluation, and management of high blood pressure in adults: a report of the American College of Cardiology/American Heart Association Task Force on Clinical Practice Guidelines. *J Am Coll Cardiol.* 2018;71:e127–e248. doi: 10.1016/j.jacc.2017.11.006
16. Grimes DA, Schulz KF. Bias and causal associations in observational research. *Lancet.* 2002;359:248–252. doi: 10.1016/S0140-6736(02)07451-2
17. Amar S, Gokce N, Morgan S, Loukideli M, Van Dyke TE, Vita JA. Periodontal disease is associated with brachial artery endothelial dysfunction and systemic inflammation. *Arterioscler Thromb Vasc Biol.* 2003;23:1245–1249. doi: 10.1161/01.ATV.0000078603.90302.4A
18. Fuchs FD, Whelton PK. High blood pressure and cardiovascular disease. *Hypertension.* 2020;75:285–292. doi: 10.1161/HYPERTENSIONAHA.119.14240
19. Sanz M, Marco Del Castillo A, Jepsen S, Gonzalez-Juanatey JR, D'Aiuto F, Bouchard P, Chapple I, Dietrich T, Gotsman I, Graziani F, et al. Periodontitis and cardiovascular diseases: consensus report. *J Clin Periodontol.* 2020;47:268–288. doi: 10.1111/jcpe.13189

20. Pietropaoli D, Del Pinto R, Ferri C, Marzo G, Giannoni M, Ortu E, Monaco A. Association between periodontal inflammation and hypertension using periodontal inflamed surface area and bleeding on probing. *J Clin Periodontol.* 2020;47:160–172. doi: 10.1111/jcpe.13216
21. Tsakos G, Sabbah W, Hingorani AD, Netuveli G, Donos N, Watt RG, D'Aiuto F. Is periodontal inflammation associated with raised blood pressure? Evidence from a National US survey. *J Hypertens.* 2010;28:2386–2393. doi: 10.1097/HJH.0b013e32833e0fe1
22. Demmer RT, Papapanou PN, Jacobs DR Jr, Desvarieux M. Bleeding on probing differentially relates to bacterial profiles: the oral Infections and vascular disease epidemiology study. *J Clin Periodontol.* 2008;35:479–486. doi: 10.1111/j.1600-051X.2008.01232.x
23. Bokhari SA, Khan AA, Butt AK, Hanif M, Izhar M, Tatakis DN, Ashfaq M. Periodontitis in coronary heart disease patients: strong association between bleeding on probing and systemic biomarkers. *J Clin Periodontol.* 2014;41:1048–1054. doi: 10.1111/jcpe.12284
24. Del Pinto R, Pietropaoli D, Munoz-Aguilera E, D'Aiuto F, Czesnikiewicz-Guzik M, Monaco A, Guzik TJ, Ferri C. Periodontitis and hypertension: is the association causal? *High Blood Press Cardiovasc Prev.* 2020;27:281–289. doi: 10.1007/s40292-020-00392-z
25. Czesnikiewicz-Guzik M, Nosalski R, Mikolajczyk TP, Vidler F, Dohnal T, Dembowska E, Graham D, Harrison DG, Guzik TJ. Th1-type immune responses to Porphyromonas gingivalis antigens exacerbate angiotensin II-dependent hypertension and vascular dysfunction. *Br J Pharmacol.* 2019;176:1922–1931.
26. Tonetti MS, D'Aiuto F, Nibali L, Donald A, Storry C, Parkar M, Suvan J, Hingorani AD, Vallance P, Deanfield J. Treatment of periodontitis and endothelial function. *N Engl J Med.* 2007;356:911–920. doi: 10.1056/NEJMoa063186
27. Desvarieux M, Demmer RT, Jacobs DR Jr, Rundek T, Boden-Albala B, Sacco RL, Papapanou PN. Periodontal bacteria and hypertension: the oral infections and vascular disease epidemiology study (INVEST). *J Hypertens.* 2010;28:1413–1421. doi: 10.1097/HJH.0b013e328338cd36
28. Libby P, Loscalzo J, Ridker PM, Farkouh ME, Hsue PY, Fuster V, Hasan AA, Amar S. Inflammation, immunity, and infection in atherothrombosis: JACC review topic of the week. *J Am Coll Cardiol.* 2018;72:2071–2081. doi: 10.1016/j.jacc.2018.08.1043
29. Arimatsu K, Yamada H, Miyazawa H, Minagawa T, Nakajima M, Ryder MI, Gotoh K, Motooka D, Nakamura S, Iida T, et al. Oral pathobiont induces systemic inflammation and metabolic changes associated with alteration of gut microbiota. *Sci Rep.* 2014;4:4828. doi: 10.1038/srep04828
30. Goh CE, Trinh P, Colombo PC, Genkinger JM, Mathema B, Uhlemann AC, LeDuc C, Leibel R, Rosenbaum M, Paster BJ, et al. Association between nitrate-reducing oral bacteria and cardiometabolic outcomes: results from ORIGINS. *J Am Heart Assoc.* 2019;8:e013324. doi: 10.1161/JAHA.119.013324
31. Tribble GD, Angelov N, Weltman R, Wang BY, Eswaran SV, Gay IC, Parthasarathy K, Dao DV, Richardson KN, Ismail NM, et al. Frequency of tongue cleaning impacts the human tongue microbiome composition and enterosalivary circulation of nitrate. *Front Cell Infect Microbiol.* 2019;9:39. doi: 10.3389/fcimb.2019.00039
32. Muñoz Aguilera E, Leira Y, Miró Catalina Q, Orlandi M, Czesnikiewicz-Guzik M, Guzik TJ, Hingorani AD, Nart J, D'Aiuto F. Is systemic inflammation a missing link between periodontitis and hypertension? Results from two large population-based surveys. *J Intern Med.* 2020. doi: 10.1111/joim.13180
33. Zhou QB, Xia WH, Ren J, Yu BB, Tong XZ, Chen YB, Chen S, Feng L, Dai J, Tao J, et al. Effect of intensive periodontal therapy on blood pressure and endothelial microparticles in patients with prehypertension and periodontitis: a randomized controlled trial. *J Periodontol.* 2017;88:711–722. doi: 10.1902/jop.2017.160447
34. Jepsen S, Blanco J, Buchalla W, Carvalho JC, Dietrich T, Dörfer C, Eaton KA, Figuero E, Frencken JE, Graziani F, et al. Prevention and control of dental caries and periodontal diseases at individual and population level: consensus report of group 3 of joint EFP/ORCA workshop on the boundaries between caries and periodontal diseases. *J Clin Periodontol.* 2017;44(suppl 18):S85–S93. doi: 10.1111/jcpe.12687
35. Choi HM, Han K, Park YG, Park JB. Associations among oral hygiene behavior and hypertension prevalence and control: the 2008 to 2010 Korea National health and nutrition examination survey. *J Periodontol.* 2015;86:866–873. doi: 10.1902/jop.2015.150025
36. Law MR, Morris JK, Wald NJ. Use of blood pressure lowering drugs in the prevention of cardiovascular disease: meta-analysis of 147 randomised trials in the context of expectations from prospective epidemiological studies. *BMJ.* 2009;338:b1665. doi: 10.1136/bmj.b1665
37. Smits KP, Listl S, Plachokova AS, Van der Galien O, Kalmus O. Effect of periodontal treatment on diabetes-related healthcare costs: a retrospective study. *BMJ Open Diabetes Res Care.* 2020;8:e001666.
38. Mahmood S, Shah KU, Khan TM, Nawaz S, Rashid H, Baqar SWA, Kamran S. Non-pharmacological management of hypertension: in the light of current research. *Ir J Med Sci.* 2019;188:437–452. doi: 10.1007/s11845-018-1889-8
39. Lindholt JS, Søgaard R. Population screening and intervention for vascular disease in Danish men (VIVA): a randomised controlled trial. *Lancet.* 2017;390:2256–2265. doi: 10.1016/S0140-6736(17)32250-X
40. Machado V, Aguilera EM, Botelho J, Basit Hussain S, Leira Y, Proença L, D'Aiuto F, João Mendes J. Association between periodontitis and high blood pressure: results from the study of periodontal health in Almada-Seixal (SoPHiAS). *J Clin Med.* 2020;9:1585.
41. Sedgwick P. Nested case-control studies: advantages and disadvantages. *BMJ : British Medical Journal.* 2014;348:g1532.
42. Gauderman WJ. Sample size requirements for matched case-control studies of gene-environment interaction. *Stat Med.* 2002;21:35–50. doi: 10.1002/sim.973
43. Pearce N. Analysis of matched case-control studies. *BMJ.* 2016;352:i969.

Periodontitis is linked to higher blood pressure values in otherwise healthy individuals



Association between periodontitis and blood pressure highlighted in systemically healthy individuals. Results from a nested case-control study. Supplementary Tables S1 and S2

Table S1: Mediation analysis: periodontal inflammation measurements (exposure), mean SBP (outcome), WBC (mediator).

Exposure definitions	WBC Mediator (Model 1)				WBC Mediator (Model 2)			
Perio vs non-perio	**Effect Estimate**	**SE**	**95% CI**	**p-value**	**Effect Estimate**	**SE**	**95% CI**	**p-value**
a (exposure → mediator)	0,561	0,146	(0,27; 0,85)	<0,001	0,561	0,146	(0,27; 0,85)	<0,001
b (mediator → outcome)	-0,00	0,38	(-0,75; 0,74)	0,994	-0,05	0,38	(-0,79; 0,70)	0,900
c (total effect)	3,361	1,26	(0,92; 5,80)	0,007	2,71	1,30	(0,16; 5,26)	0,037
c' (direct effect)	3,363	1,26	(0,88; 5,84)	0,008	2,73	1,32	(0,15; 5,32)	0,038
ab (mediated effect)	-0,00	1,21	(-0,42; 0,42)	0,994	-0,03	0,21	(-0,44; 0,39)	0,900
ab/c(percentage mediated) = %	0,00	0,06	(-0,12; 0,12)	0,994	-0,01	0,08	(-0,16; 0,14)	0,899
Mean PPD	**Effect Estimate**	**SE**	**95% CI**	**p-value**	**Effect Estimate**	**SE**	**95% CI**	**p-value**
a (exposure → mediator)	0,199	0,06	(0,08; 0,32)	0,001	0,199	0,06	(0,08; 0,32)	0,001
b (mediator → outcome)	-0,027	0,38	(-0,76; 0,71)	0,943	-0,067	0,38	(-0,80; 0,67)	0,859
c (total effect)	1,857	0,51	(0,85; 2,86)	<0,001	1,577	0,55	(0,50; 2,65)	0,004
c' (direct effect)	1,862	0,52	(0,84; 2,81)	<0,001	1,590	0,55	(0,50; 2,67)	0,004
ab (mediated effect)	-0,005	0,07	(-0,15; 0,14)	0,943	-0,013	0,07	(-0,16; 0,13)	0,860
ab/c(percentage mediated) = %	-0,000	0,04	(-0,08: 0,08)	0,942	-0,008	0,05	(-0,10; 0,08)	0,859

Mean CAL	Effect Estimate	SE	95% CI	p-value	Effect Estimate	SE	95% CI	p-value
a (exposure → mediator)	0,144	0,05	(0,05; 0,24)	0,003	0,144	0,05	(0,05; 0,24)	0,003
b (mediator → outcome)	-0,008	0,38	(-0,74; 0,73)	0,982	-0,047	0,038	(-0,78; 0,69)	0,902
c (total effect)	1,512	0,41	(0,69; 2,32)	<0,001	1,270	0,45	(0,39; 2,15)	0,005
c' (direct effect)	1,513	0,42	(0,69; 2,33)	<0,001	1,277	0,45	(0,39; 2,16)	0,005
ab (mediated effect)	-0,001	0,05	(-0,11; 0,10)	0,982	-0,007	0,05	(-0,11; 0,10)	0,902
ab/c(percentage mediated) = %	-0,001	0,03	(-0,07; 0,07)	0,982	-0,005	0,04	(-0,08; 0,08)	0,901
Mean FMBS	**Effect Estimate**	**SE**	**95% CI**	**p-value**	**Effect Estimate**	**SE**	**95% CI**	**p-value**
a (exposure → mediator)	0,010	0,00	(0,00; 0,01)	<0,001	0,010	0,00	(0,00; 0,01)	<0,001
b (mediator → outcome)	-0,074	0,38	(-0,81; 0,66)	0,844	-0,134	0,38	(-0,87; 0,60)	0,721
c (total effect)	0,097	0,02	(0,05; 0,14)	<0,001	0,088	0,02	(0,04; 0,13)	<0,001
c' (direct effect)	0,097	0,02	(0,05; 0,14)	<0,001	0,089	0,02	(0,04; 0,14)	<0,001
ab (mediated effect)	-0,001	0,00	(-0,01; 0,01)	0,844	-0,001	0,00	(-0,01; 0,01)	0,722
ab/c(percentage mediated) = %	-0,01	0,04	(0,01; 0,06)	0,843	-0,014	0,04	(-0,09; 0,06)	0,719

FMBS, Full mouth gingival bleeding score; CAL, Clinical attachment level; CI, Confidence interval; PPD, Probing pocket depth; SBP, Systolic blood pressure; SE, Standard error, and WBC, White blood cells.
Model 1: Unadjusted
Model 2: Adjusted for Age, BMI, gender, ethnicity, smoking, physical activity, family history of CVD

Table S2: Mediation analysis: periodontal inflammation measurements (exposure), SBP ≥140 mm Hg (outcome), WBC (mediator).

Exposure definitions	WBC Mediator (Model 1)				WBC Mediator (Model 2)			
Perio vs non-perio	**Effect Estimate**	**SE**	**95% CI**	**p-value**	**Effect Estimate**	**SE**	**95% CI**	**p-value**
a (exposure → mediator)	0,561	0,14	(0,27; 0,85)	<0,001	0,561	0,15	(0,27; 0,85)	<0,001
b (mediator → outcome)	-0,003	0,01	(-0,02; 0,01)	0,688	-0,003	0,01	(-0,01; 0,01)	0,713
c (total effect)	0,064	0,03	(0,01; 0,12)	0,020	0,068	0,03	(0,01; 0,12)	0,018
c' (direct effect)	0,066	0,03	(0,01; 0,12)	0,019	0,070	0,03	(0,01; 0,13)	0,017
ab (mediated effect)	-0,002	0,00	(-0,01; 0,01)	0,690	-0,002	0,00	(-0,01; 0,01)	0,714
ab/c(percentage mediated) = %	-0,03	0,07	(-0,17; 0,11)	0,686	-0,025	0,07	(-0,16; 0,11)	0,711
Mean PPD	**Effect Estimate**	**SE**	**95% CI**	**p-value**	**Effect Estimate**	**SE**	**95% CI**	**p-value**
a (exposure → mediator)	0,199	0,06	(0,08; 0,32)	0,001	0,199	0,06	(0,08; 0,04)	0,001
b (mediator → outcome)	-0,004	0,01	(-0,02; 0,01)	0,604	-0,004	0,01	(-0,02; 0,01)	0,648
c (total effect)	0,040	0,01	(0,02; 0,06)	<0,001	0,044	0,01	(0,02; 0,07)	<0,001
c' (direct effect)	0,041	0,01	(0,02; 0,06)	<0,001	0,044	0,01	(0,02; 0,07)	<0,001
ab (mediated effect)	-0,001	0,00	(-0,00; 0,00)	0,608	-0,001	0,00	(-0,00; 0,00)	0,651
ab/c(percentage mediated) = %	-0,021	0,04	(-0,10; 0,06)	0,605	-0,02	0,04	(-0,09; 0,06)	0,648

Mean CAL	Effect Estimate	SE	95% CI	p-value	Effect Estimate	SE	95% CI	p-value
a (exposure → mediator)	0,144	0,05	(0,05; 0,24)	0,003	0,144	0,05	(0,05; 0,24)	0,003
b (mediator → outcome)	-0,004	0,01	(-0,02; 0,01)	0,641	-0,00	0,01	(-0,20; 0,01)	0,691
c (total effect)	0,032	0,01	(0,01; 0,05)	<0,001	0,036	0,01	(0,02; 0,05)	<0,001
c' (direct effect)	0,033	0,01	(0,01; 0,05)	<0,001	0,036	0,01	(0,02; 0,06)	<0,001
ab (mediated effect)	-0,001	0,00	(-0,00; 0,00)	0,645	0,000	0,00	(-0,00; 0,00)	0,694
ab/c(percentage mediated) = %	-0,017	0,04	(-0,09; 0,05)	0,642	-0,013	0,03	(-0,08; 0,05)	0,692
Mean FMBS	**Effect Estimate**	**SE**	**95% CI**	**p-value**	**Effect Estimate**	**SE**	**95% CI**	**p-value**
a (exposure → mediator)	0,010	0,00	(0,00; 0,01)	<0,001	0,010	0,00	(0,00; 0,01)	<0,001
b (mediator → outcome)	-0,005	0,01	(-0,02; 0,01)	0,563	-0,005	0,01	(-0,02; 0,01)	0,586
c (total effect)	0,002	0,00	(0,00; 0,00)	<0,001	0,002	0,00	(0,00; 0,00)	<0,001
c' (direct effect)	0,002	0,00	(0,00; 0,00)	<0,001	0,002	0,00	(0,00; 0,00)	<0,001
ab (mediated effect)	0,000	0,00	(0,00; 0,00)	0,568	0,000	0,00	(0,00; 0,00)	0,590
ab/c(percentage mediated) = %	-0,024	0,04	(-0,11; 0,06)	0,563	-0,022	0,04	(-0,10; 0,06)	0,586

FMBS, Full mouth gingival bleeding score; CAL, Clinical attachment level; CI, Confidence interval; PPD, Probing pocket depth; SBP, Systolic blood pressure; SE, Standard error, and WBC, White blood cells.
Model 1: Unadjusted.
Model 2: Adjusted for Age, BMI, gender, ethnicity, smoking, physical activity, family history of CVD

Lecture for federal university of Paraná (Brasil)

Brazilian Ministry of Education
FEDERAL UNIVERSITY OF PARANÁ
DEPARTMENT OF STOMATOLOGY
SECTION OF PERIODONTOLOGY

Dr. Eva Muñoz Aguilera
Periodontology
265 Grays Inn Road
London
London
WC1X 8LD

May 14th, 2020.

Dear Dr. Muñoz-Aguilera,

Thank you for joining our online Seminar entitled "*Hypertension and Periodontitis*" as a **Lecturer** on May 12th, 2020 (1 hour). The Seminar is a part of our "Special Topics in Periodontology" Course at the Postgraduate Programme in Dentistry at The Federal University of Paraná. Our students and I really appreciate your effort and collaboration.

Sincerely,

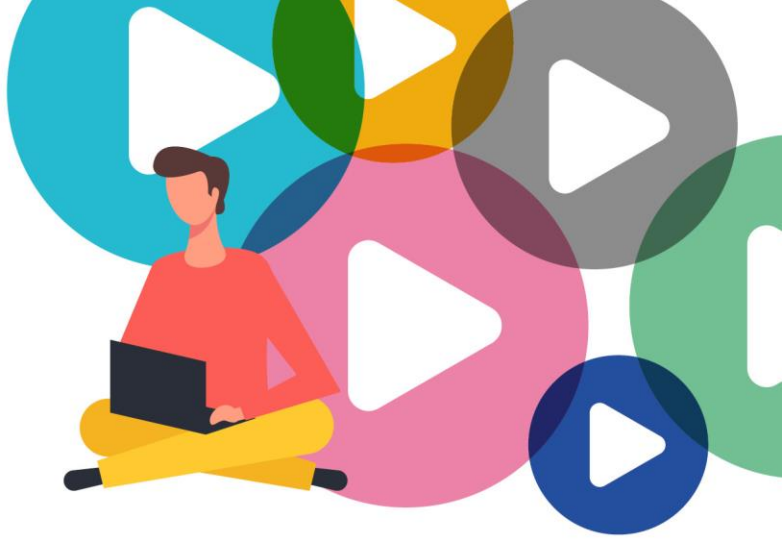
Dr. Joao Paulo Steffens
Organizer

Lecture for SEPA: Alianza por la Salud Oral y General

Sepa'20 OnAir

Un congreso para todos

SepaTV



C E R T I F I C A D O
P O N E N T E

La Sociedad Española de Periodoncia y Osteointegración
CERTIFICA QUE:

Eva Muñoz

Ha impartido la sesión

El efecto del tratamiento periodontal no quirúrgico sobre la HTA.

En el Congreso Sepa OnAir 2020, celebrado de forma virtual entre el 11 Septiembre y el 28 Noviembre de 2020.

Antonio Bujaldón
Presidente de SEPA

Sepa.

SepaTV