Oxidative Stress and Antioxidants in the Diagnosis and Therapy of Periodontitis

L’ubomíra Tóthová 1,2 and Peter Celec 1,3,4*

1 Faculty of Medicine, Institute of Molecular Biomedicine, Comenius University, Bratislava, Slovakia, 2 Faculty of Medicine, Institute of Physiology, Comenius University, Bratislava, Slovakia, 3 Faculty of Medicine, Institute of Pathophysiology, Comenius University, Bratislava, Slovakia, 4 Department of Molecular Biology, Faculty of Natural Sciences, Comenius University, Bratislava, Slovakia

Oxidative stress has been implicated in the pathogenesis of numerous diseases. However, large interventional studies with antioxidants failed to show benefits in the prevention or treatment of cardiovascular diseases, cancer, or diabetes mellitus. Numerous clinical studies have confirmed the association of oxidative stress markers and periodontitis. Technical and biological variability is high for most of the analyzed markers and none of them seems to be optimal for routine clinical use. In a research setting, analysis of a palette of oxidative stress markers is needed to cover lipid peroxidation, protein oxidation, and the antioxidant status. The source of reactive oxygen species and their role in the pathogenesis of periodontitis remains unclear. Intervventional experiments indicate that oxidative stress might be more than just a simple consequence of the inflammation. Small studies have confirmed that some antioxidants could have therapeutic value at least as an addition to the standard non-surgical treatment of periodontitis. A clear evidence for the efficiency of antioxidant treatment in large patient cohorts is lacking. Potentially, because lowering of oxidative stress markers might be a secondary effect of anti-inflammatory or antibacterial agents. As the field of research of oxidative stress in periodontitis gains attraction and the number of relevant published papers is increasing a systematic overview of the conducted observational and interventional studies is needed. This review summarizes the currently available literature linking oxidative stress and periodontitis and points toward the potential of adjuvant antioxidant treatment, especially in cases where standard treatment fails to improve the periodontal status.

Keywords: reactive oxygen species, free radicals, antioxidative therapy, systematic review, oral diseases

INTRODUCTION

Oxidative stress is both, a pathomechanism involved in numerous inflammatory diseases causing damage to lipids, nucleic acids and proteins—oxidative distress, as well as an important physiological process that enables the immune system to cope with microorganisms and intracellular cell signaling—oxidative eustress (Sies et al., 2017). Which edge of the sword is the dominant depends on the delicate balance between the production of reactive oxygen/nitrogen species and the antioxidant capacity of the tissue. The physiological functions of free radicals have
been neglected for years and so, much more is known about the pathological role of oxidative stress. A variety of free radicals is produced and interacts with a variety of substrates. This leads to a palette of biomarkers that can be used for the assessment of oxidative stress-induced damage (Frijhoff et al., 2015).

Oxidative stress is usually defined as a disbalance of the production of free radicals and antioxidant mechanisms (Kopáni et al., 2006). However, free radicals are not a simple negative byproduct of oxygen metabolism. They are involved in immune responses, liver metabolism, but also in intracellular signaling pathways (Espíñosa-Diez et al., 2015). The mechanisms involved in the physiological intracellular role of free radicals include the modulation of cysteine residues of redox-sensitive enzymes and other regulatory proteins (Finkel, 2011; Russell and Cotter, 2015). It has been hypothesized that under physiological conditions even high concentrations of any one primary reactive oxygen or nitrogen species does not lead to oxidative damage, as the cell has preventive and reparative tools to cope with the radical. The reactions of superoxide, nitric oxide, and other primary reactive species are reversible and are ideal for intracellular signaling (Weidinger and Kozlov, 2015). Thus, measuring oxidative stress using any one marker can lead to wrong interpretations. This might include the hope that administration of antioxidants will effectively treat diseases associated with oxidative stress. As recently hypothesized, one of the prototypic oxidative stress diseases—diabetes mellitus might be actually a consequence of deficiency of reactive oxygen species rather than of oxidative damage (Watson, 2014). It should not escape our notice that this proposed mechanism might suggest that antioxidants could increase the risk rather than prevent metabolic diseases.

Periodontitis is an inflammatory disease affecting supporting structures of the teeth leading at the end to loss of alveolar bone and teeth (Kinane et al., 2017). The main causative factor are microorganisms that colonize the subgingival dental plaque inducing an inflammatory host response. The inflammation affects, however, also the surrounding healthy tissue ultimately leading to the destruction of the periodontium (Kinane et al., 2011). Although lipopolysaccharide and proteolytic enzymes are essential in periodontitis, exaggerated inflammatory response, genetic predisposition, smoking, bad oral hygiene, and malnutrition are important in pathogenesis of periodontitis as well (Laine et al., 2012).

The role of oxidative stress in periodontitis has been postulated already decades ago (Shapira et al., 1991; Chapple, 1997). However, the suggested involvement was not clear. Some studies showed that leukocytes from patients with periodontitis are exhausted and have a low oxidation activity (Loesche et al., 1988), other studies pointed toward higher production of free radicals by leukocytes from periodontitis patients (Kimura et al., 1993). The contradictory findings from these studies might be related to the dynamics of the mechanisms during the pathogenesis of the disease, but they might also be explained by the different forms of periodontitis (Biasi et al., 1999).

The term oxidative stress is vague, similarly, antioxidants might affect many processes not directly related to free radical generation or action (Niki, 2016). This makes the interpretation of studies focusing on oxidative stress in periodontitis difficult. A systematic review of these clinical studies and animal experiments might, thus, be needed.

**BIOMARKERS OF OXIDATIVE STRESS AND PATHOGENESIS OF PERIODONTITIS**

Surprisingly, many observational studies analyzing oxidative stress in patients with periodontitis had relatively consistent results with higher oxidative stress markers in either saliva or blood and/or decreased antioxidant status in comparison to controls. The summary table of the identified studies can be found in Table 1. One of the largest observational studies has shown that the antioxidant status in blood, analyzed as vitamin C, bilirubin, and calculated total antioxidant capacity was inversely associated with mild and severe periodontitis (Chapple et al., 2007b). The more severe periodontitis, the clearer the association. Additionally, in a subgroup of never-smokers the antioxidants seemed to protect against development of periodontitis. Others smaller studies confirmed these results. Total antioxidant capacity was lower in plasma or serum of patients with chronic periodontitis (Chapple et al., 2002; Brock et al., 2004; Konopka et al., 2007). Similarly, superoxide dismutase activity (Huang et al., 2014) along with catalase and glutathione peroxidase activity (Tonguç et al., 2011) as important contributor to the total antioxidant capacity was found to be lower in periodontitis. In line with these results from blood, in the majority of studies, the antioxidant status was lower also locally—in saliva. The total antioxidant status/potential/capacity is in general the ability of a tissue to resist artificially induced oxidative stress, but studies differ in the analytical approaches. Nevertheless, the local antioxidant capacity was found to be lower in saliva from patients with an aggressive form of periodontitis when compared to chronic periodontitis (Acquier et al., 2017). Patients with chronic periodontitis have lower antioxidant capacity than control patients (Zhang et al., 2015; Ahmad-Motamayel et al., 2017). Although the total antioxidant capacity is not a specific marker of antioxidant power, uric acid, glutathione peroxidase (Miricescu et al., 2014), and reduced glutathione (Gumus et al., 2009) as specific antioxidants were reported to be significantly lower in saliva of patients with chronic or aggressive periodontitis. On contrary, activities of the major antioxidant enzymes were found to be higher in chronic periodontitis patients in all investigated samples, i.e., plasma, erythrocytes, and in the gingival tissue (Panjamurthy et al., 2005). Similarly, SOD2 and GPX1 genes were overexpressed in the gingiva of chronic periodontitis patients (Duarte et al., 2012). Lactoferrin, myeloperoxidase and interleukin 1 beta were all positively correlated with the clinical markers of periodontal damage (Wei et al., 2004). However, whether such associations of higher antioxidant and pro-inflammatory response are a consequence or cause of severe periodontitis cannot be judged only from observations.

Regarding the analyzed markers of oxidative stress, a comparison of the published studies is complicated, if not impossible due to the huge variability of measured markers.
| Study design       | Sample size | Note                  | Sample type | Outcome                                                                 | References               |
|-------------------|-------------|-----------------------|-------------|--------------------------------------------------------------------------|--------------------------|
| Cross-sectional study | 20 ApG patients | —                     | Saliva      | ↑ ROS, TBARS with ApG; TRAP ↓ with ApG compared to ChP                     | Acquier et al., 2017     |
|                   | 20 ChP patients |                       | Serum       | ↓ TAC in serum and saliva; ↓ MDA increased in serum and saliva vs. controls | Ahmadi-Motamayel et al., 2017 |
|                   | 20 controls   |                       | Saliva      | ↑ TAC, SOD in PW vs. non-PW and in ChP group compared to controls; ↑ TAC, SOD in third trimester in PW with ChP vs. PW with ChP in the first trimester | Akalin et al., 2009      |
| Case-control study (cross-sectional) | 55 ChP patients |                       | Serum       | ↑ plasma small molecule antioxidant capacity                              | Allen et al., 2011       |
|                   | 55 healthy controls |                       | GCF         | ↑ PC in T2DM with periodontitis vs. PH T2DM patients                        | Almerich-Silla et al., 2015 |
| Prospective study | 33 PW with ChP | Pregnancy             | Serum       | ↓ TAC, SOD activity in post-menopausal women with ChP in serum and GCF     | Baltacioglu et al., 2006 |
|                   | 18 PW with gingivitis |                       | GCF         | ↓ TAC, SOD activity in post-menopausal women with ChP in serum and GCF     | Baltacioglu et al., 2014 |
|                   | 21 PW controls  |                       | GCF         | ↓ TAC in GCF and plasma in ChP vs. control; ↓ TAC in saliva in ChP vs. control | Banasová et al., 2015    |
|                   | 27 non-PW ChP  |                       | GCF         | ↓ TAC in GCF and plasma in ChP vs. control; ↓ TAC in saliva in ChP vs. control | Borges et al., 2007      |
|                   | 25 non-PW controls |                       | GCF         | ↓ TAC in GCF and plasma in ChP vs. control; ↓ TAC in saliva in ChP vs. control | Brock et al., 2004       |
| Comparative study (cross-sectional) | 20 T2DM patients with periodontitis |                       | T2DM Plasma | ↓ plasma small molecule antioxidant capacity                              | Acquier et al., 2017     |
|                   | 20 T2DM patients PH |                       | T2DM Plasma | ↑ PC in T2DM with periodontitis vs. PH T2DM patients                        | Almerich-Silla et al., 2015 |
| Cross-sectional study | 33 ChP patients | —                     | Saliva      | ↑ TAC, 8-OHdG, MDA and activity of SOD, GPx in ChP vs. controls            | Almerich-Silla et al., 2015 |
|                   | 16 patients with gingivitis |                       | Saliva      | ↑ TAC, 8-OHdG, MDA and activity of SOD, GPx in ChP vs. controls            | Almerich-Silla et al., 2015 |
|                   | 37 healthy controls |                       | Saliva      | ↑ TAC, 8-OHdG, MDA and activity of SOD, GPx in ChP vs. controls            | Almerich-Silla et al., 2015 |
| Observational study (cross-sectional) | 19 non-T2DM patients |                       | T2DM Saliva | ↓ GPx, glutathione reductase in poor metabolic control T2DM patients; ↑ GSSG/GSH ratio in poor metabolic control T2DM | Arana et al., 2017       |
|                   | 24 T2DM patients with good metabolic control |                       | T2DM Saliva | ↓ GPx, glutathione reductase in poor metabolic control T2DM patients; ↑ GSSG/GSH ratio in poor metabolic control T2DM | Arana et al., 2017       |
|                   | 27 T2DM patients with poor metabolic control T2DM |                       | T2DM Saliva | ↓ GPx, glutathione reductase in poor metabolic control T2DM patients; ↑ GSSG/GSH ratio in poor metabolic control T2DM | Arana et al., 2017       |
| Case-control study | 15 PH + normal weight | Obesity               | GCF         | ↑ MDA, PC and ↓ TAC in ChP + obese group                                   | Atalay et al., 2017      |
|                   | 15 gingivitis + normal weight |                      | GCF         | ↑ MDA, PC and ↓ TAC in ChP + obese group                                   | Atalay et al., 2017      |
|                   | 15 ChP + normal weight |                      | GCF         | ↑ MDA, PC and ↓ TAC in ChP + obese group                                   | Atalay et al., 2017      |
|                   | 15 PH + obese     |                      | GCF         | ↑ MDA, PC and ↓ TAC in ChP + obese group                                   | Atalay et al., 2017      |
|                   | 18 gingivitis + obese |                      | GCF         | ↑ MDA, PC and ↓ TAC in ChP + obese group                                   | Atalay et al., 2017      |
|                   | 15 ChP + obese    |                      | GCF         | ↑ MDA, PC and ↓ TAC in ChP + obese group                                   | Atalay et al., 2017      |
| Cross-sectional study | 32 ChP post-menopausal women | Menopause         | Serum GCF   | ↑ TAC, SOD activity in post-menopausal women with ChP in serum and GCF     | Baltacioglu et al., 2006 |
|                   | 31 ChP pre-menopausal |                      | Serum GCF   | ↑ TAC, SOD activity in post-menopausal women with ChP in serum and GCF     | Baltacioglu et al., 2006 |
|                   | 25 PH post-menopausal women |                    | Serum GCF   | ↑ TAC, SOD activity in post-menopausal women with ChP in serum and GCF     | Baltacioglu et al., 2006 |
|                   | 26 PH pre-menopausal women |                    | Serum GCF   | ↑ TAC, SOD activity in post-menopausal women with ChP in serum and GCF     | Baltacioglu et al., 2006 |
| Cross-sectional study | 35 ApG patients | —                     | Serum       | ↑ MDA, TOS, OSI in periodontitis groups; ↓ TAC in periodontitis groups vs. controls (except serum MDA) | Baltacioglu et al., 2014 |
|                   | 33 ChP patients | —                     | Serum       | ↑ MDA, TOS, OSI in periodontitis groups; ↓ TAC in periodontitis groups vs. controls (except serum MDA) | Baltacioglu et al., 2014 |
|                   | 30 PH controls  | —                     | Serum       | ↑ MDA, TOS, OSI in periodontitis groups; ↓ TAC in periodontitis groups vs. controls (except serum MDA) | Baltacioglu et al., 2014 |
| Cross-sectional study | 23 ChP patients | —                     | Saliva      | ↑ TBARS in ChP (men); ↓ TAC in ChP (women); trend toward ↓ DNA integrity in ChP | Banasová et al., 2015     |
|                   | 19 PH controls  | —                     | Saliva      | ↑ TBARS in ChP (men); ↓ TAC in ChP (women); trend toward ↓ DNA integrity in ChP | Banasová et al., 2015     |
| Cross-sectional study | 9 patients with periodontitis | —                     | Gingival tissue | ↑ activities of MPO, GPx, glutathione S-transferase in periodontitis group; ↑ TBARS and GSSG levels increased vs. controls | Borges et al., 2007       |
|                   | 9 healthy controls | —                     | Gingival tissue | ↑ activities of MPO, GPx, glutathione S-transferase in periodontitis group; ↑ TBARS and GSSG levels increased vs. controls | Borges et al., 2007       |
| Cross-sectional study | 20 ChP patients 20 healthy controls | —                     | Serum Saliva GCF | ↑ TAC in GCF and plasma in ChP vs. control; ↓ TAC in saliva in ChP vs. control | Brock et al., 2004       |
| Cross-sectional study | 32 ChP patients | —                     | Saliva      | ↑ 8-OHdG and mtDNA deletions in ChP group vs. control                     | Canakci et al., 2009     |
|                   | 32 PH control   | —                     | Saliva      | ↑ 8-OHdG and mtDNA deletions in ChP group vs. control                     | Canakci et al., 2009     |
| Cross-sectional study | 30 ChP patients | —                     | Blood       | ↑ mtDNA deletion in ChP group vs. control                                  | Canakci et al., 2006     |
|                   | 30 PH control   | —                     | Blood       | ↑ mtDNA deletion in ChP group vs. control                                  | Canakci et al., 2006     |
| Cross-sectional study | 10 preeclampsia ChP | Preeclampsia       | Serum Saliva GCF | ↓ TAC in ChP women with preeclampsia (GCF, serum, saliva); ↑ MDA in ChP preeclamptic women (GCF, serum) | Canakci et al., 2007     |
|                   | 10 preeclampsia PH |                        | Serum Saliva GCF | ↓ TAC in ChP women with preeclampsia (GCF, serum, saliva); ↑ MDA in ChP preeclamptic women (GCF, serum) | Canakci et al., 2007     |
|                   | 10 normotensive ChP |                        | Serum Saliva GCF | ↓ TAC in ChP women with preeclampsia (GCF, serum, saliva); ↑ MDA in ChP preeclamptic women (GCF, serum) | Canakci et al., 2007     |
|                   | 10 normotensive PH |                        | Serum Saliva GCF | ↓ TAC in ChP women with preeclampsia (GCF, serum, saliva); ↑ MDA in ChP preeclamptic women (GCF, serum) | Canakci et al., 2007     |
| Cross-sectional study | 31 pre-menopausal | Menopause             | GCF         | ↑ 8-OHdG in ChP post-menopausal women in GCF and gingival tissue           | Chandra et al., 2017     |
|                   | 31 peri-menopausal |                      | GCF         | ↑ 8-OHdG in ChP post-menopausal women in GCF and gingival tissue           | Chandra et al., 2017     |
|                   | 31 post-menopausal ChP women |            | GCF         | ↑ 8-OHdG in ChP post-menopausal women in GCF and gingival tissue           | Chandra et al., 2017     |
| Cross-sectional study | 10 ChP patients | —                     | Plasma      | ↓ TAC in GCF and plasma in ChP vs. control; ↓ GSH and GSSG in GCF in ChP | Chapple et al., 2002     |
|                   | 10 PH control   | —                     | Plasma      | ↓ TAC in GCF and plasma in ChP vs. control; ↓ GSH and GSSG in GCF in ChP | Chapple et al., 2002     |

(Continued)
| Study design               | Sample size                                             | Note                           | Sample type     | Outcome                                                                 | References          |
|---------------------------|----------------------------------------------------------|--------------------------------|-----------------|-------------------------------------------------------------------------|---------------------|
| Observational correlational study | 11,480 participants; 1,567 with mild periodontitis 609 with severe periodontitis |                                | Serum           | ↓ vitamin C, bilirubin and TAC (calculated) in mild or severe periodontitis | Chapple et al., 2007b |
| Cross-sectional study     | 17 periodontitis patients 20 healthy controls            |                                | Saliva          | ↓ TAC in periodontitis                                                  | Diab-Lacki et al., 2003 |
| Cross-sectional study     | 12 SH and PH controls 8 well-controlled T2DM patients with ChP 14 poor-controlled T2DM patients with ChP | T2DM                           | Gingival tissue | Peroxiredoxin 1 and GPX1 overexpressed in ChP; Peroxiredoxin 2 and SOD2 up-regulated especially in poor—controlled T2DM with ChP | Duarte et al., 2012 |
| Cross-sectional study     | 20 ChP patients with RA 20 PH patients with RA 20 ChP patients SH 20 SH and PH controls | Rheumatoid arthritis Serum GCF |                  | ↑ TOS in GCF of ChP and RA ChP groups (no difference for TOS and OSI in serum) | Eisen et al., 2012  |
| Cross-sectional study     | 18 ChP patients with hyperlipidemia 18 PH patients with hyperlipidemia 19 CHP patients SH 19 PH SH controls | Hyperlipidemia                  | Serum           | ↑ MDA and 8-OHdG in patients with ChP and hyperlipidemia               | Fentoglu Ö et al., 2015 |
| Cross-sectional study     | 24 ChP with depression 23 CHP without depression          | Depression                      | Plasma          | ↑ nitric oxide metabolites, lipid peroxides, AOPP and TRAP in CHP with depression | Gomes et al., 2017  |
| Cross-sectional study     | 16 T1DM with periodontitis 25 T2DM with periodontitis 24 SH with periodontitis | T1DM                           | Saliva          | ↓ GSH and ↓ GSsO in the patients with T1DM                             | Gurus et al., 2009  |
| Case-control study        | 115 P women (6 month postpartum follow-up) 72 non-P women | Pregnancy                      | Saliva          | ↑ 8-OHdG in PW; ↓ GPx decreased in PW; ↓ TBARS postpartum vs. non-pregnant women | Gümüş et al., 2015  |
| Prospective study         | 218 P women 459 P women with mild periodontitis 114 P women with moderate-severe periodontitis | Pregnancy                      | Serum           | ↑ 8-isoprostane in PW with moderate-severe periodontitis               | Hickman et al., 2011 |
| Prospective study         | 50 ChP patients 50 healthy controls                      |                                | Serum           | ↓ SOD in saliva and serum; ↑ prostaglandin E2, D2, prostaglandin F2α and TXB2, 5- hydroxyeicosatetraenoic acid, F2-isoprostanes; ↑ prostacyclin I2 and 13- hydroxyoctadecadienoic acid, 9- hydroxyoctadecadienoic acid | Huang et al., 2014  |
| Cross-sectional study     | 55 patients with DS 74 patients with mental retardation 88 healthy controls | Down Syndrome                  | Whole blood     | ↑ oxidative burst activity of blood (monocytes and granulocytes) in DS patients with decreased periodontal health | Khocht et al., 2014  |
| Cross-sectional study     | 26 AgP patients 30 CHP patients 25 healthy controls      |                                | Gingival blood  | ↑ 8-OHdG in gingival blood of CHP and AgP patients; ↓ TAC in gingival blood of CHP patients; ↓ TAC in peripheral blood of both groups |
| Cross-sectional study     | 1,258 old men                                            | Old age                        | Serum           | ↓ beta-cryptoxanthin and beta-carotene with decreased periodontal health of old men | Linden et al., 2009  |
| Cross-sectional study     | 356 periodontitis patients 207 PH controls               |                                | Plasma          | ↑ reactive oxygen metabolites and shorter leukocyte telomere length in CHP | Masi et al., 2011  |
| Cross-sectional study     | 20 CHP patients 20 PH controls                            |                                | Saliva          | ↑ 8-OHdG, MDA in CHP; ↓ uric acid, GPx activities and TAC in CHP (correlation with bone loss markers) | Miricescu et al., 2014 |
| Cross-sectional study     | 10 T2DM patients PH 8 SH controls                         | T2DM                           | Periodontal tissue | ↑ MDA, ↓ GSH in periodontal tissue of T2DM                             | Monea et al., 2014  |
| Cross-sectional study     | 24 CHP patients with ACS 24 patients PH with ACS 24 CHP patients without ACS 24 controls PH without ACS | Acute coronary syndrome        | Saliva          | ↑ 8-OHdG, MDA, and PC in patients (correlation with periodontal and cardiovascular markers) | Nguyen et al., 2016  |

(Continued)
### TABLE 1 | Continued

| Study design            | Sample size                                      | Note | Sample type          | Outcome                                                                 | References                                   |
|-------------------------|--------------------------------------------------|------|----------------------|--------------------------------------------------------------------------|----------------------------------------------|
| Cross-sectional study   | 25 ChP patients                                  | –    | Plasma               | ↑ TBARS in ChP; † SOD, CAT, GPx activities in ChP; ↓ vitamins E, C and GSH in ChP | Panjamurthy et al., 2005                     |
|                         | 25 healthy controls                              |      | Gingival tissue      |                                                                          |                                              |
|                         |                                                  |      | Erythrocytes         |                                                                          |                                              |
| Cross-sectional study   | 29 ChP patients                                  | –    | Saliva               | ↑ 8-OHdG in ChP (correlation with P gingivalis)                          | Sawamoto et al., 2005                        |
|                         | 20 healthy controls                              |      |                      |                                                                          |                                              |
| Cross-sectional cohort study | 46 severe periodontitis patients                   | –    | Saliva               | ↑ PC increased in severe periodontitis; ↓ urate and FRAP in severe periodontitis | Sculey and Langley-Evans, 2003                |
|                         | 37 moderate periodontitis patients                |      |                      |                                                                          |                                              |
|                         | 48 mild periodontitis and healthy                 |      |                      |                                                                          |                                              |
| Cross-sectional study   | 20 ChP patients with RA                           |      | Rheumatoid arthritis | ↑ OSI and prolidase in ChP patients with RA                               | Sezer et al., 2013                            |
|                         | 20 PH patients with RA                            |      |                      |                                                                          |                                              |
|                         | 20 ChP patients without RA                        |      |                      |                                                                          |                                              |
|                         | 20 PH SH controls                                 |      |                      |                                                                          |                                              |
| Cross-sectional study   | 20 ChP patients                                  | –    | Psoriasis            | ↑ OSI (irrespective of periodontitis) in patients groups; (Ps and PsA showed no effect on clinical parameters in ChP patients) | Sezer et al., 2016                          |
|                         | 20 PH patients with Ps                              |      |                      |                                                                          |                                              |
|                         | 20 PH patients with PsA                           |      |                      |                                                                          |                                              |
|                         | 20 PH patients with PsA                           |      |                      |                                                                          |                                              |
| Cross-sectional study   | 4,717 participants                                | Diabetes mellitus | Serum                | Periodontitis with highest 8-isoprostane quartile associated with ↑ CRP | Singer et al., 2015                          |
|                         |                                                  | Hypertension       |                      |                                                                          |                                              |
| Cross-sectional study   | 29 severe periodontitis                           | Diabetes mellitus | Serum                | ↑ ROM in patients with worst periodontal status                          | Tamaki et al., 2014b                          |
|                         | 77 moderate periodontitis                         | Hypertension       |                      |                                                                          |                                              |
|                         | 96 mild periodontitis and healthy                 |      |                      |                                                                          |                                              |
| Cross-sectional study   | 25 severe periodontitis                           | Diabetes mellitus | Saliva               | Superoxide and hydroxyl radical scavenging activities associated with periodontitis | Tamaki et al., 2015                            |
|                         | 43 moderate periodontitis                         | Hypertension       |                      |                                                                          |                                              |
|                         | 92 mild periodontitis and healthy                 |      |                      |                                                                          |                                              |
| Cross-sectional study   | 39 patients with periodontitis                    | –    | Saliva               | ↓ antioxidant concentrations related to periodontal status               | Tartaglia et al., 2017                        |
| Cross-sectional study   | 23 smokers with ChP                              | Smoking           | Serum                | ↑ MDA in serum and gingival tissue in smoking Chp patients groups; ↓ SOD, CAT and GPx in Chp groups | Tonguç et al., 2011                          |
|                         | 23 former smokers with ChP                       |      |                      |                                                                          |                                              |
|                         | 19 non-smokers with ChP                          |      |                      |                                                                          |                                              |
|                         | 20 PH non-smokers controls                        |      |                      |                                                                          |                                              |
| Cross-sectional study   | 82 children                                      | –    | Saliva               | TBARS in correlation to periodontal status; TAC related to periodontal status and oral hygiene; AOPP related to caries in children | Tothova et al., 2013                          |
| Cross-sectional study   | 30 ChP patients with T2DM                        | T2DM             | Plasma               | ↑ MDA in Chp patients irrespective of T2DM                               | Trivedi et al., 2014                          |
|                         | 30 ChP SH patients                               |      | Saliva               |                                                                          |                                              |
|                         | 30 PH patients with T2DM                         |      | Red blood cell lyse |                                                                          |                                              |
|                         | 30 PH SH controls                                |      |                      |                                                                          |                                              |
| Cross-sectional study   | 100 ChP patients                                 | –    | Saliva               | ↑ 8-OHdG and human neutrophil elastase/alpha1-proteinase inhibitor in ChP | Villa-Correa et al., 2015                     |
|                         | 50 healthy controls                              |      |                      |                                                                          |                                              |
| Cross-sectional study   | 19 patients with periodontitis                    | –    | GCF                  | ↑ GPx, lactoferrin, myeloperoxidase and IL-1beta in periodontal tissues (correlation with clinical periodontal markers) | Wei et al., 2004                             |
|                         | 8 healthy controls                               |      |                      |                                                                          |                                              |
| Cross-sectional study   | 31 smokers                                       | Smoking           | Saliva               | ↑ 8-epi-PGF(2alpha) with periodontal status and smoking; Smoking ↑ TXB(2) and PGF(2alphas) and ↓ 6-oxo-PGF(1alpha) | Wolfram et al., 2006                          |
|                         | 90 non-smokers                                   |      |                      |                                                                          |                                              |
| Cross-sectional study   | 58 ChP patients                                  | –    | Saliva               | ↑ micronuclei and nuclear abnormalities, as well as 8-OHdG in both periodontitis groups | Zamora-Perez et al., 2015                     |
|                         | 42 AgP patients                                  |      | Buccal mucosa        |                                                                          |                                              |
|                         | 60 healthy controls                              |      |                      |                                                                          |                                              |
| Cross-sectional study   | 45 severe periodontitis patients                  | –    | Saliva               | ↓ TAC in periodontitis (TOS no difference)                               | Zhang et al., 2015                            |
|                         | 37 healthy controls                              |      |                      |                                                                          |                                              |

**ChP:** chronic periodontitis; **AgP:** aggressive periodontitis; **SH:** systematically healthy; **PH:** periodontal healthy; **T2DM:** type 1 diabetes mellitus; **T2DM:** type 2 diabetes mellitus; **Ps:** psoriasis; **PsA:** psoriatic arthritis; **DS:** Down syndrome; **PW:** pregnant woman; **RA:** Rheumatoid arthritis; **ACS:** Acute coronary syndrome; **GCF:** gingival crevicular fluid; **TAC:** total antioxidant capacity; **TBARS:** thiobarbituric acid reacting substances; **8-HdG:** 8-hydroxydeoxyguanosine; **GPx:** glutathione peroxidase; **SOD:** superoxide dismutase; **OSI:** oxidative stress index; **IL-1beta:** interleukin 1beta; **ROM:** reactive oxygen metabolites; **ROS:** reactive oxygen species; **FRAP:** ferric reducing antioxidant power; **4-HNE:** 4-hydroxy-2-nonenal; **CAT:** catalase; **CRP:** C-reactive protein; **AOPP:** advanced oxidation protein products; **TRAP:** total radical-trapping antioxidant potential; **MPO:** myeloperoxidase; **mtDNA:** mitochondrial DNA.
Malondialdehyde, 8-hydroxydeoxyguanosine (Konopka et al., 2007; Almerich-Silla et al., 2015), protein carbonyls (Nguyen et al., 2016; Atabay et al., 2017), thiobarbituric acid reacting substances (Borges et al., 2007), nitric oxide, advanced oxidation protein products, lipid peroxidation products (Gomes et al., 2017), 8-isoprostanes (Hickman et al., 2011) were all higher in patients with periodontitis. The most commonly measured markers of oxidative stress seem to be malondialdehyde and thiobarbituric acid reacting substances pointing toward oxidative damage of lipids, especially, lipid membranes. Lipid peroxidation was higher in saliva (Tóthova et al., 2013), serum (Tonguç et al., 2011), and in the gingival tissue (Panjamurthy et al., 2005) of patients with chronic or aggressive periodontitis. Correlational studies confirmed a positive association of these markers with periodontal status scores (Chapple et al., 2007b; Tamaki et al., 2014b). The less commonly measured markers related to oxidative damage as mitochondrial DNA (Canakci et al., 2006), micronuclei and nuclear abnormalities (Zamora-Perez et al., 2015), as well as a leukocyte telomere length shortening (Masi et al., 2011) were all higher in periodontitis as well.

Oxidative stress was found to be involved in the pathogenesis of many diseases besides periodontitis. Virtually, almost all inflammatory diseases lead to increased oxidative stress. This in turn can trigger more damage to the tissues, not excluding the gingival tissue and, thus, worsening periodontitis. There are several studies describing oxidative stress in systemic diseases with regard to the periodontal status. Nguyen et al. (2016) investigated patients with the acute coronary syndrome with or without chronic periodontitis, patients with periodontitis and healthy controls. Lipid, protein, and DNA oxidation markers were higher in periodontitis than in the control group (Nguyen et al., 2016). Another study with rheumatoid arthritis and chronic periodontitis confirmed higher oxidative stress markers in plasma and lower antioxidant capacity in both groups when compared to healthy control. However, if both comorbidities were present, there was no further enhancement of oxidative stress (Sezer et al., 2013).

Taken together, observational cross-sectional studies confirmed the association of oxidative stress and periodontitis. Higher oxidative stress and lower antioxidant status can be detected in plasma, saliva as well as in the gingival crevicular fluid of patients with various clinical forms of periodontitis. These findings support the use of body fluids, but especially the non-invasive diagnostic fluid saliva, as suitable sample types for diagnostics or monitoring the course of periodontitis. Current data do not support the use of a single oxidative stress marker. It is likely that a set of markers covering both, oxidative damage and antioxidative status, will be needed. The low specificity of oxidative stress markers calls for caution when interpreting the results even if more than one marker is used. The inter-individual and intra-individual variability of the analyzed markers is very high. This prevents their use at the level of individual diagnostics. The overview of the published observational studies shows the enormous heterogeneity of the patient populations as well as the used analytical tools. In a meta-analysis focusing on systematic oxidative stress, it was shown that higher malondialdehyde and nitric oxide as well as lower total antioxidant capacity of plasma/serum characterize patients with periodontitis in comparison to controls (Liu et al., 2014). Based on our overview, taken into account systemic and local oral biomarkers of oxidative stress, it is clear that there is a need for both, the use of a wider palette of markers to analyze oxidative stress and its causes in more detail, and the introduction of new biomarkers with a better sensitivity/specificity profile in specific subgroups of patients. Of special importance is the small sample size in most studies. A collaborative effort with a multi-center recruitment of patients and a standardized consensus protocol in the pre-analytical and analytical phase is highly needed.

**OXIDATIVE STRESS AND TREATMENT OF PERIODONTITIS**

The implication of oxidative stress in the pathogenesis of cardiovascular diseases and cancer as the major causes of death in the combination with the widespread availability of dietary antioxidants started a hype that was supported by the hypothesis that aging is caused by oxidative stress (Harman, 1956; Finkel and Holbrook, 2000). However, the hype was quickly over as large clinical studies revealed that antioxidants or at least antioxidant vitamins were not able to prevent any of the diseases of aging (Coulter et al., 2006; Sesso et al., 2008; Myung et al., 2013). Some studies even revealed a slight but increased risk in patients taking antioxidants in preeclampsia (Rumbold et al., 2006) and lung cancer (Alpha-Tocopherol, 1994). One meta-analysis showed that taking antioxidant may even increase the all-cause mortality by 5% (Bjelakovic et al., 2007). This might be related to the terminological and mechanistic confusion about antioxidants, which might have a very indirect effect on the production or effects of reactive oxygen species. In an era of evidence-based medicine the clear conclusion is that antioxidant dietary supplements should undergo clinical evaluation before marketing similarly to other medicinal drugs (Bjelakovic et al., 2012). Recent mechanistic experiments shed light on the details how antioxidants may stimulate tumor growth (Sayin et al., 2014) or increase the risk of metastasis (Piskounova et al., 2015). Antioxidants are highly variable in their mechanism and structure. Diseases and patients vary even more. So, negative results from oncology or cardiology should not be generalized to other diseases including periodontitis. It is very likely that the role of oxidative stress changes during disease progression and, thus, the potential antioxidant treatment affecting not only oxidative damage but also the inflammatory process might have different affects at various stages of the complex pathogenesis. The issue of antioxidants that mostly do not affect free radicals and their action *in vivo*, but rather interfere with variable cellular signaling pathways has been reviewed in a recently published paper (Azzi, 2017).

Many published studies have analyzed the effects of periodontitis treatment on oxidative stress (Table 2). Studies of special interest are those with covariates or comorbidities that were taken into account. A trial by Guentisch et al. (2008) examined healthy subjects and patients with periodontitis further.
### TABLE 2 | Interventional studies analyzing the effect of periodontitis on oxidative stress.

| Treatment | Treatment duration | Sample size | Comorbidity | Sample type | Results | References |
|-----------|--------------------|-------------|-------------|-------------|---------|------------|
| NST with vitamin C (2,000 mg/day) | 1 month | 30 ChP patients | None | Plasma | ↓ TAC in ChP patients; NST lead to ↑ TAC in ChP patients; No effect of vitamin C | Abou Sulaiman and Shehadeh, 2010 |
| NST with lycopene (8 mg/day) | 2 month | 20 ChP patients | None | Serum | ↓ MDA and clinical parameters after therapy | Ambati et al., 2017 |
| Oral hygiene education with insulin treatment for T1DM | 3 months | 32 T1DM patients at diagnosis<br>18 SH children with gingivitis<br>18 SH and PH children | T1DM | Serum, Saliva, GCF | ↑ serum, salivary and GCF OSI in T1DM group; ↓ after treatment | Aral et al., 2017 |
| NST with lycopene (8 mg/day) | – | 42 ChP patients | None | Plasma | ↑ IL-1ss, UA and clinical parameters after treatment | Arora et al., 2013 |
| NST | 14 days | 13 ChP patients with FMF<br>15 ChP SH patients<br>14 PH patients with FMF<br>15 PH and SH controls | Familial Mediterranean fever | Serum, GGF | ↓ periodontal clinical markers after treatment; ↓ TOS in GGF in FMF patients with ChP; no difference in serum TOS and OSI | Bostanci et al., 2014 |
| ST with antioxidant gel (2mg lycopene) | 1 week | 31 ChP patients | None | GCF | ↓ 8-OHdG after treatment, as well as periodontal clinical markers | Chandra et al., 2013 |
| NST | 2 months | 35 ChP patients<br>32 healthy controls | None | Plasma, GCF | ↓ TAC in GCF in ChP; ↑ after treatment; (plasma TAC no difference at baseline between groups, no change after treatment) | D’Aauto et al., 2010 |
| NST | 4 h | 145 ChP patients (14 with therapy)<br>56 healthy controls | None | Serum | Patients with severe periodontitis ↑ROM ↓ TAC; ↑ ROM after treatment | Dominguez et al., 2010 |
| NST with fluorescence-controlled Er:YAG laser radiation | Laser therapy applied 1 day after NST | 30 ChP patients | None | GCF | no difference between with/without laser treatment in TAC and clinical parameters; IL-1beta and ↑ TNF-alpha after NST only; ↓ after NST with laser | D’Aauto et al., 2010 |
| NST | – | 30 ChP patients<br>30 healthy controls | Smokers/non-smokers | Serum, Saliva | ↑ MDA in smoking ChP patients; ↑ GPx in ChP groups; ↓ TAC in ChP groups; ↓ MDA and GPx after therapy | Guentsch et al., 2008 |
| NST | 14 days | 47 (24 / 23) ChP patients<br>46 (23/23) healthy controls | Smokers/non-smokers | Serum, Saliva | ↑ salivary 8-OHdG and GPx in ChP; ↑ 4-HNE in GCF in ChP smokers; ↑ 8-OHdG after therapy in saliva and GCF | Hendek et al., 2015 |
| NST | – | 7 ChP patients<br>7 healthy controls | None | Saliva | ↑ TAC and ↓ SOD activity in ChP; ↓ TAC, SOD activity immediately after NST | Kim et al., 2010 |
| NST | – | 60 moderate to severe periodontitis patients with T2DM | T2DM | Serum | No effect of NST on d-8-iso, MMP-2, MMP-9 and hsCRP | Koromantzos et al., 2012 |
| NST; NST with antioxidants (6 mg/day; lycopene, zinc, and selenium) or antioxidants only | 3 doses in 2 weeks | 30 ChP patients<br>30 gingivitis patients<br>10 healthy controls | None | Saliva | ↓ UA in ChP patients; antioxidant treatment ↑ UA | Mathur et al., 2013 |
| Tai Chi (5 days a week, 60 min) | 6 months | 71 sedentary patients with periodontitis | Old age | Saliva | ↑ TAC, SOD after therapy | Mendoza-Núñez et al., 2014 |
| NST and surgical treatment | Surgical treatment 6 weeks after NST | 12 AgP patients | None | Serum, Plasma | Periodontitis severity associated with LDL concentrations; No changes in lipid profile after treatment; No difference in GSH and lipid hyperoxide after therapy | Nibali et al., 2015 |
TABLE 2 | Continued

| Treatment | Treatment duration | Sample size | Comorbidity | Sample type | Results | References |
|-----------|--------------------|-------------|-------------|-------------|---------|------------|
| NST or oral hygiene instructions | 2 visits within 7 days | 42 ChP patients | None | Saliva | ↑ TAC, ALB, UA, GPx and ↓ SOD; (no effect of oral hygiene instructed therapy was found) | Novakovic et al., 2014 |
| NST | – | 25 severe ChP patients | None | Serum | Saliva | ↑ 8-OHdG | Onder et al., 2017 |
| NST with Coenzyme Q10; NST with tea tree oil gel or placebo | 7 days | 15 ChP patients (moderate to severe) | None | - | Both antioxidant treatment procedures effective in ↓ clinical markers of ChP (PI, GI, PPD and CAL) | Raut and Sethi, 2016 |
| NST and oral hygiene instructions | 2–4 months | 29 ChP patients | None | Saliva | ↑ 8-OHdG in ChP patients before therapy; ↓ 8-OHdG and P. gingivalis after treatment, as well as periodontal clinical markers | Sawamoto et al., 2005 |
| NST | – | 8 ChP patients | None | Platelet suspension | ↓ periodontal clinical parameters and CRP after therapy, as well as ↑ cGMP and SOD activity | Siqueira et al., 2013 |
| Surgical treatment with taurine (500 mg/day) | 15 days | 10 ChP patients | None | Plasma Gingival tissue | ↓ TBARS, GPx in plasma and gingival tissue; ↑ GSH and ↓ periodontal clinical parameters after therapy | Sree and Sethupathy, 2014 |
| NST | – | 78 ChP patients | None | Saliva | ↑ 8-OHdG in ChP patients before therapy; ↓ 8-OHdG and periodontal clinical markers after therapy | Takane et al., 2002 |
| NST | – | 22 ChP patients | None | Plasma | ↑ oxidative index, oxidized LDL and CRP in ChP; ↓ of these parameters after treatment | Tamaki et al., 2011 |
| NST | – | 25 ChP patients | None | Serum | ↓ serum TAC and CAT in both groups of patients; TAC ↑ in ChP patients after therapy (no treatment effect on CAT) | Thomas et al., 2014 |
| NST | 4 visits within 14 days | 30 (15/15) ChP patients | Smokers/non-smokers | GCF | ↑ IL-1beta in ChP patients; ↓ IL-1beta after NST irrespective of smoking; (no difference in TAC and TOS before or after treatment between groups) | Toker et al., 2012 |
| NST | 2 weeks | 25 ChP patients with MS | Metabolic syndrome | Serum | TOS and OSI showed no difference between groups in serum after therapy; ↑ TAC of MS ChP patients before treatment, but ↓ after therapy in serum; ↓ OSI and ↑ TAC in both groups after treatment in salvia | Torumtay et al., 2016 |
| NST | Once per week/1 month | 22 ChP patients | None | Saliva | ↑ SOD in patients with low dental visits after NST; ↑ TAC in patients with regular dental visits after therapy | Yang et al., 2014 |
| NST with dietary intervention | 3 visits/6 months | 37 ChP patients (19 without intervention; 18 with intervention) | None | Plasma | ↑ TAC after dietary intervention in plasma; no differences in periodontal clinical parameters after dietary intervention | Zare Javid et al., 2014 |

CHP, chronic periodontitis; SH, systematically healthy; PH, periodontal healthy; NST, non-surgical treatment; ST, surgical treatment; T1DM, type 1 diabetes mellitus; T2DM, type 2 diabetes mellitus; FMF, familial Mediterranean fever; MS, metabolic syndrome; GCF, gingival crevicular fluid; TAC, total antioxidant capacity; TBARS, thiobarbituric acid reacting substances; 8-HOdG, 8-hydroxydeoxyguanosine; GPx, glutathione peroxidase; GSH, reduced glutathione; MDA, malondialdehyde; TOS, total oxidant status; SOD, superoxide dismutase; OSI, oxidative stress index; IL-1ss, salivary interleukin 1beta; ROM, reactive oxygen metabolites; 4-HNE, 4-hydroxy-2-nonenal; hsCRP, high-sensitivity C-reactive protein; d-8-iso, d-8-iso prostaglandin F2α; MMP-2, matrix metalloproteinase 2; MMP-9, matrix metalloproteinase 9; LDL, low density lipoprotein; ALB, albumin; UA, uric acid; CAT, catalase; CRP, C-reactive protein; PI, plaque index; GI, gingival bleeding index; PPD, probing pocket depth; CAL, clinical attachment level; TNF-alpha, tumor necrosis factor alpha; cGMP, L-arginine-nitric oxide (NO)-cyclic guanosine monophosphate.
divided into smokers and non-smokers. While smokers with periodontitis displayed highest malondialdehyde concentration and highest glutathione peroxidase activity along with lowest total antioxidant capacity in saliva. The non-surgical treatment helped to normalize the values regardless of the smoking status (Guenthsch et al., 2008). Several other trials confirmed the consistency of these findings (Chapple et al., 2007a; Abou Sulaiman and Shehadeh, 2010; Hendek et al., 2015). On the other hand, a severe systemic condition such as type 2 diabetes may lead to inefficiency of non-surgical therapies to improve the periodontal status or oxidative stress (Koromantzos et al., 2012). In general, most of the studies clearly show an improvement of oxidative damage after standard treatment of periodontitis. The studies, however, vary greatly regarding sample types, markers measured, and most cohorts were very small and highly variable. Thus, making relevant conclusions or recommendations for the clinical dentistry is difficult, if not impossible. Future research efforts should focus on the lack of uniformity and standardization as well as on the issue of the low informative value of small patient cohorts.

Although the causality of the association between oxidative stress and periodontitis is everything but clear, some clinical studies already tested antioxidants in periodontitis. One small, placebo controlled, randomized, and double-blind study showed, that single application of lycopene gel to periodontitis patients improved clinical attachment and decreased oxidative stress in gingival crevicular fluid (Chandra et al., 2013). Nevertheless, a published systematic review on antioxidant treatment of periodontitis revealed that a consistent effect in randomized clinical trials was found only for lipophile antioxidants such as lycopene and vitamin E, but not for hydrophile antioxidants such as vitamin C (Muniz et al., 2015). This might be related to the vulnerability of lipids to oxidative damage, but also to mitochondria as the site of effect of some antioxidants. Interestingly, antioxidants targeting directly mitochondria have been shown to be effective in decreasing inflammatory activity and organ damage in animal model of sepsis (Lowes et al., 2013).

ANIMAL EXPERIMENTS

The high number of observational and interventional studies analyzing the association between oxidative stress and periodontitis indicates that there are many open questions that cannot be answered by more and more clinical studies. Many of the questions need controlled conditions in experiments. The number of animal experiments analyzing the role oxidative stress in periodontitis is small, but it increases. Periodontitis is mostly induced by ligature placement around the first molars of the animals or local injection of periodontal pathogens or their toxins (Genco et al., 1998; Fine, 2009; Oz and Puleo, 2011). In such a rat model, it was shown that periodontitis leads to an increased production of reactive oxygen species and markers of oxidative damage (Ekuni et al., 2010). In addition, oxidative stress induced by periodontitis seems to be associated with the dynamics and severity of the periodontal inflammation (Bosca et al., 2016).

Some animal experiments focus on the distant effects of periodontitis that seem to be mediated by oxidative stress. It was shown that mitochondrial DNA is oxidatively modified in the liver, kidney, heart, and brain of rats with induced periodontitis (Tomofuji et al., 2011). Another similar experiment by the same group has shown that periodontitis might worsen ethanol-induced liver damage (Tomofuji et al., 2008). The oxidative damage to the heart, but also endothelial dysfunction and resulting atherosclerosis induced by experimental periodontitis can be prevented by antioxidant treatment (Ekuni et al., 2009; Ozdem et al., 2017; Saito et al., 2017). A majority of the published experiments focus on the use of antioxidants such as vitamin C (Tomofuji et al., 2009b), N-acetylcysteine (Toker et al., 2009), or resveratrol (Tamaki et al., 2014a), but also drugs with an antioxidant activity beyond their main mechanism of action (de Araujo Junior et al., 2013; Culic et al., 2014; Oktay et al., 2015). Dietary interventions have also been investigated—high cholesterol diet seems to worsen periodontitis (Tomofuji et al., 2006). On contrary, the phytoestrogen genistein and capro-enriched diet was shown to be protective against periodontal damage and oxidative stress induced by periodontitis in mice and rats, respectively (Tomofuji et al., 2009a; Bhattarai et al., 2017). The most promising candidate drug at least according to animal experiments is melatonin. This amphiphile molecule has an optimal distribution in the tissues and can, thus, reach the periodontal tissues even after systemic administration (Köse et al., 2017). Of special clinical relevance is the induction of periodontitis in diabetic rats since periodontitis is a common complication of diabetes in humans. Melatonin was able to prevent alveolar bone loss also in this experimental model (Köse et al., 2016). However, regarding the mechanism of action, it is not clear whether the protective effect of melatonin is due to its direct antioxidant characteristics or due to its immunomodulatory effects that might reduce oxidative stress indirectly (Kara et al., 2013). This uncertainty is not specific for melatonin. Any antioxidant might affect the immune response and, thus, have anti-inflammatory properties. Beyond established systemic antioxidants novel approaches with a local periodontal application are tested (Saita et al., 2016). Experimental tools such as genetically engineered mice that produce luciferase under the regulation of transcription factors related to oxidative stress and antioxidant response have been developed and might greatly improve our understanding of the role of oxidative stress in periodontitis (Kataoka et al., 2016). The new treatment options together with new and improved models could be very helpful in the fight against this widespread and serious disease.

CONCLUSION

The role of oxidative stress in periodontitis is not clear despite decades of research. Numerous studies have been published showing the potential of oxidative stress markers for screening, diagnosis or monitoring of the disease, but none is in routine clinical use. Similarly, animal experiments, as well as most of
the interventional studies in patients, indicate that antioxidant treatment should be effective in the therapy of periodontitis, but no such treatment has been approved. The lack of translation could be either due to the lack of strong evidence for the clinical usefulness or due to obstacles in the application of the results including the low or absent commercial interest from major stakeholders. From a research perspective, an important issue is the lack of specificity—both, in diagnostics and treatment. Not even the source of free radical production is clear. While some studies point toward neutrophils (Katsuragi et al., 2003), others show that bacteria actively producing reactive oxygen species might contribute to oxidative stress in periodontitis (Huycke et al., 2002; Vlkova and Celec, 2009). It is of crucial importance that the number of conducted animal experiments in this field is increasing, especially of those focusing on the dynamics of oxidative stress during disease progression. The antioxidant treatment might be effective only in a subset of patients during a specific stage of periodontitis. The shift from pure observations to interventions and animal experiments that can be followed in the published literature in the recent years is highly positive and should bring this field of research closer to true clinical applications despite chronic lack of financial support and human resources.

**AUTHOR CONTRIBUTIONS**

L’T has analyzed the data from the literature, prepared tables, and drafted the manuscript; PC has designed the review, conducted the literature search, and drafted the manuscript.

**FUNDING**

The authors are supported from the European Operational Program funded by the ERDF (project ITMS: 26240120027).

**REFERENCES**

Abou Sulaiman, A. E., and Shehadeh, R. M. (2010). Assessment of total antioxidant capacity and the use of vitamin C in the treatment of non-smokers with chronic periodontitis. *J. Periodontol.* 81, 1547–1554. doi: 10.1902/jop.2010.100173

Acquier, A. B., De Couto Pita, A. K., Busch, L., and Sánchez, G. A. (2017). Parameters of oxidative stress in saliva from patients with aggressive and chronic periodontitis. *Redox Rep.* 22, 119–126. doi: 10.1080/13510002.2016.1198104

Ahmadi-Motamayez, F., Goodarzi, M. T., Jamshidi, Z., and Kebræiæ, R. (2017). Evaluation of salivary and serum antioxidant and oxidative stress statuses in patients with chronic periodontitis: a case-control study. *Front. Physiol.* 8:189. doi: 10.3389/fphys.2017.00189

Akalin, F. A., Baltacioglu, E., Alver, A., and Karabulut, E. (2009). Total antioxidant capacity and superoxide dismutase activity levels in serum and gingival crevicular fluid in pregnant women with chronic periodontitis. *J. Periodontol.* 80, 457–467. doi: 10.1902/jop.2009.080218

Allen, E. M., Matthews, J. B., O’Halloran, D. J., Griffiths, H. R., and Chapple, I. L. (2011). Oxidative and inflammatory status in Type 2 diabetes patients with periodontitis. *J. Clin. Periodontol.* 38, 894–901. doi: 10.1111/j.1600-051X.2011.01764.x

Almerich-Silla, J. M., Montiel-Company, J. M., Pastor, S., Serrano, F., Puig-Silla, M., and Dasi, F. (2015). Oxidative stress parameters in saliva and its association with periodontal disease and types of bacteria. *Dis. Markers* 2015:653537. doi: 10.1155/2015/653537

Alpha-Tocopherol, Beta Carotene Cancer Prevention Study Group. (1994). The effect of vitamin E and beta carotene on the incidence of lung cancer and other cancers in male smokers. *N. Engl. J. Med.* 330, 1029–1035. doi: 10.1056/NEJM19940113301501

Ambati, M., Rani, K. R., Reddy, P. V., Suryaprassana, J., Dasari, R., and Gireddy, H. (2017). Evaluation of oxidative stress in chronic periodontitis patients following systemic antioxidant supplementation: a clinical and biochemical study. *J. Nat. Sci. Biol. Med.* 8, 99–103. doi: 10.1016/j.jsbmb.2011.01764.x

Aral, C. A., Nalbantoglu O. O., Nur, B. G., Altunsoy, M., and Aral, K. (2017). Metabolic control and periodontal treatment decreases elevated oxidative stress in the early phases of type 1 diabetes onset. *Arch. Oral Biol.* 82, 115–120. doi: 10.1016/j.archoralbio.2017.06.009

Arana, C., Moreno-Fernández, A. M., Gómez-Moreno, G., Morales-Portillo, C., Serrano-Olmedo, I., de la Cuesta Mayor, M. C., et al. (2017). Increased salivary oxidative stress parameters in patients with type 2 diabetes: relation with periodontal disease. *Endocr. Diabetes Nutr.* 64, 258–264. doi: 10.1016/j.endinu.2017.03.005

Arora, N., Avula, H., and Avula, J. K. (2013). The adjunctive use of systemic antioxidant therapy (lycopene) in nonsurgical treatment of chronic periodontitis: a short-term evaluation. *Quintessence Int.* 44, 395–405. doi: 10.3290/j.qi.a29188

Atabay, V. E., Lutfioglu, M., Avci, B., Sakallioglu, E. E., and Aydögu, A. (2017). Obesity and oxidative stress in patients with different periodontal status: a case-control study. *J. Periodontol.* 52, 51–60. doi: 10.1111/jre.12368

Aziz, A. (2017). Antioxidants: wonder drugs or quackery? *Biofactors*. doi: 10.1002/biof.1388. [Epub ahead of print].

Baltacioglu, E., Akalin, F. A., Alver, A., Balaban, F., Unsal, M., and Karabulut, E. (2006). Total antioxidant capacity and superoxide dismutase activity levels in serum and gingival crevicular fluid in post-menopausal women with chronic periodontitis. *J. Clin. Periodontol.* 33, 385–392. doi: 10.1111/j.1600-051X.2006.00923.x

Baltacioglu, E., Yuva, P., Aydin, G., Alver, A., Kahraman, C., Karabulut, E., et al. (2014). Lipid peroxidation levels and total oxidant/antioxidant status in serum and saliva from patients with chronic and aggressive periodontitis. oxidative stress index: a new biomarker for periodontal disease? *J. Periodontol.* 85, 1432–1441. doi: 10.1902/jop.2014.130654

Banasová, L., Kamodyová, N., Jansáková, K., Tóthová, L., Stanko, P., Turna, J., et al. (2015). Salivary DNA and markers of oxidative stress in patients with chronic periodontitis. *Clin. Oral Investig.* 19, 201–207. doi: 10.1007/s00784-014-1236-z

Bhattarai, G., Poudel, S. B., Kook, S. H., and Lee, J. C. (2017). Anti-inflammatory, anti-osteoclastic, and antioxidant activities of genistein protect against alveolar bone loss and periodontal tissue degradation in a mouse model of periodontitis. *J. Biomed. Mater. Res. A* 105, 2510–2521. doi: 10.1002/jbma.a.36109

Biasi, D., Bambara, L. M., Carletto, A., Caramaschi, P., Andrioli, G., Urbani, G., et al. (1999). Neutrophil migration, oxidative metabolism and adhesion in early onset periodontitis. *J. Clin. Periodontol.* 26, 563–568. doi: 10.3109/016005199.1999.260901.x

Bjelakovic, G., Nikolova, D., Gud, L. L., Simonetti, R. G., and Gud, C. (2007). Mortality in randomized trials of antioxidant supplements for primary and secondary prevention: systematic review and meta-analysis. *JAMA* 297, 842–857. doi: 10.1001/jama.297.8.842

Bjelakovic, G., Nikolova, D., Gud, L. L., Simonetti, R. G., and Gud, C. (2012). Antioxidant supplements for prevention of mortality in healthy participants and patients with various diseases. *Cochrane Database Syst. Rev.* Cd007176. doi: 10.1002/14651858.CD007176.pub2

Borges, I. Jr., Moreira, E. A., Filho, D. W., de Oliveira, T. B., da Silva, M. B., and Fröde, T. S. (2007). Neutrophil migration, oxidative metabolism and adhesion in early onset periodontitis. *J. Clin. Periodontol.* 33, 385–392. doi: 10.1111/j.1600-051X.2006.00923.x

Boča, A. B., Milčuš, V., Ilea, A., Cămpian, R. S., Rus, V., Ruxanda, F., et al. (2016). Role of nitro-oxidative stress in the pathogenesis of experimental rat periodontitis. *Clujul. Med.* 89, 150–159. doi: 10.15386/cjmed-529
Tóthová and Celec Oxidative Stress and Periodontitis

Canakci, C. F., Canakci, V., Cicek, Y., and Canakci, H. (2007). Total antioxidant capacity and antioxidant enzymes in serum, saliva, and gingival crevicular fluid of preeclamptic women with and without periodontal disease. J. Periodontol. 78, 1602–1611. doi: 10.1902/jop.2007.060046

Chandra, R. V., Sailaja, S., and Reddy, A. A. (2017). Estimation of tissue and crevicular fluid oxidative stress marker in premenopausal, premenopausal and postmenopausal women with chronic periodontitis. Gerodontology 34, 382–389. doi: 10.1111/ger.12279

Chandra, R. V., Srinivas, G., Reddy, A. A., Reddy, B. H., Reddy, C., Nagarajan, S., et al. (2013). Locally delivered antioxidant gel as an adjunct to nonsurgical therapy improves measures of oxidative stress and periodontal disease. J. Periodontal Implant Sci. 43, 121–129. doi: 10.5051/jpis.2013.43.3.121

Chapple, I. L. (1997). Reactive oxygen species and antioxidants in inflammatory diseases. J. Clin. Periodontol. 24, 287–296. doi: 10.1111/j.1600-051X.1997.tb00760.x

Chapple, I. L., Brock, G. R., Milward, M. R., Ling, N., and Matthews, J. B. (2007a). Increased salivary level of 8-hydroxydeoxyguanosine is a marker of premature oxidative mitochondrial DNA damage in gingival tissue of patients with periodontitis. Arch. Immunol. Ther. Exp. 57, 205–211. doi: 10.1007/s00005-009-0026-9

Ekuni, D., Endo, Y., Irie, K., Azuma, T., Tamaki, N., Tomofuji, T., et al. (2010). Imbalance of oxidative/anti-oxidative stress induced by periodontitis is involved in apoptosis of rat submandibular glands. Arch. Oral Biol. 55, 170–176. doi: 10.1016/j.archoralbio.2009.11.013

Esen, C., Alkan, B. A., Kirmaz, M., Akgül, O., Işıkoglu, S., and Erol, O. (2012). The effects of chronic periodontitis and rheumatoid arthritis on serum and gingival crevicular fluid total antioxidant/oxidant status and oxidative stress index. J. Periodontol. 83, 773–779. doi: 10.1902/jop.2011.110420

Espinoza-Diez, C., Miguel, V., Menenicher, D., Kietzmann, T., Sánchez-Pérez, P., Cadenas, S., et al. (2015). Antioxidant responses and cellular adjustments to oxidative stress. Redox Biol. 6, 183–197. doi: 10.1016/j.redox.2015.07.008

Fengtou, Ö. O., Kürzuoglu, F. Y., Bulut, M. T., Kumbul Doguç, D., Kulaç, E., Önder, C., et al. (2015). Evaluation of lipid peroxidation and oxidative DNA damage in patients with periodontitis and hyperlipidemia. J. Periodontol. 86, 682–688. doi: 10.1902/jop.2015.140561

Fine, D. H. (2009). Of mice and men: animal models of human periodontal disease. J. Clin. Periodontol. 36, 913–914. doi: 10.1111/j.1600-051X.2009.01456.x

Finkel, T. (2011). Signal transduction by reactive oxygen species. J. Cell Biol. 194, 7–15. doi: 10.1083/jcb.201102095

Finkel, T., and Holbrook, N. J. (2000). Oxidants, oxidative stress and the biology of aging. Nature 408, 239–247. doi: 10.1038/35041687

Frijhoff, J., Winyard, P. G., Zarkovic, N., Davies, S. S., Stocker, R., Cheng, D., et al. (2015). Clinical relevance of biomarkers of oxidative stress. Antioxid. Redox Sign. 23, 1144–1170. doi: 10.1089/ars.2015.6317

Genco, C. A., Van Dyke, T., and Aamar, S. (1998). Animal models for Porphyromonas gingivalis-mediated periodontal disease. Trends Microbiol. 6, 444–449. doi: 10.1016/S0966-842X(98)01363-8

Gomes, C., Martinho, F. C., Barbosa, D. S., Antunes, L. S., Póvoa, H. C. C., Baltus, T. H. H., et al. (2017). Increased root canal endotoxin levels are associated with chronic apical periodontitis, increased oxidative and nitrosative stress, major depression, severity of depression, and a lowered quality of life. Mol. Neurobiol. doi: 10.1007/s12035-017-0545-z. [Epub ahead of print]

Guetsch, A., Preshaw, P., Bremer-Streek, S., Klinger, G., Glockmann, E., and Sigusch, B. (2008). Lipid peroxidation and antioxidant activity in saliva of periodontitis patients: effect of smoking and periodontal treatment. Clin. Oral Investig. 12, 345–352. doi: 10.1007/s00784-008-0202-z

Gümüş, P., Buduneli, N., Cetinkalp, S., Hawkins, S. I., Renaud, D., Kinane, D. F., et al. (2009). Salivary antioxidants in patients with type 1 or 2 diabetes and inflammatory periodontal disease: a case-control study. J. Periodontol. 80, 1440–1446. doi: 10.1902/jop.2009.0900159

Gümüş, P., Emrini, G., Özaltın, V. O., Belbasakis, G. N., and Bostanci, N. (2015). Oxidative stress markers in saliva and periodontal disease status: modulation during pregnancy and postpartum. BMC Infect. Dis. 15:261. doi: 10.1186/s12879-015-0103-z

Harman, D. (1956). Aging: a theory based on free radical and radiation chemistry. J. Gerontol. 11, 298–300. doi: 10.1093/geronj/11.3.298

Hendek, M. K., Erdemir, E. O., Kisa, U., and Ozcan, G. (2015). Effect of initial periodontal therapy on oxidative stress markers in gingival crevicular fluid, saliva, and serum in smokers and non-smokers with chronic periodontitis. J. Periodontol. 86, 273–282. doi: 10.1902/jop.2014.140338

Hickman, M. A., Boggs, K. A., Moss, K. L., Beck, J. D., and Offenbacher, S. (2011). Maternal periodontal disease is associated with oxidative stress during pregnancy. Am. J. Perinatol. 28, 247–252. doi: 10.1055/s-0030-1208706

Huang, Y., Zhu, M., Li, Z., Sa, R., Chu, Q., Zhang, Q., et al. (2014). Mass spectrometry-based metabolomic profiling identifies alterations in salivary redox status and fatty acid metabolism in response to inflammation and oxidative stress in periodontal disease. Free Radic. Biol. Med. 70, 223–232. doi: 10.1016/j.freeradbiomed.2014.02.024

Huyeck, M. M., Abrams, V., and Moors, D. R. (2002). Enterococcus faecalis produces extracellular superoxide and hydrogen peroxide that damages colonic epithelial cell DNA. Carcinogenesis 23, 529–536. doi: 10.1093/carcin/23.3.529

Kara, A., Akman, S., Ozkantar, S., Tozoglu, U., Kalkan, Y., Canalci, C. F., et al. (2013). Immune modulatory and antioxidant effects of melatonin
in experimental periodontitis in rats. *Free Radic. Biol. Med.* 55, 21–26. doi: 10.1016/j.freeradbiomed.2012.11.002

Kataoka, K., Ekuni, D., Tomofuji, T., Irie, K., Kunitomo, M., Uchida, Y., et al. (2016). Visualization of oxidative stress induced by experimental periodontitis in keap1-dependent oxidative stress detector-luciferase mice. *Int. J. Mol. Sci.* 17:1907. doi: 10.3390/ijms17101907

Katsuragi, H., Ohtake, M., Kurasawa, I., and Saito, K. (2003). Intracellular production and extracellular release of oxygen radicals by PMNs and oxidative stress on PMNs during phagocytosis of periodontopathic bacteria. *Odontology* 91, 13–18. doi: 10.1007/s10266-003-0022-1

Khoct, A., Russell, B., Cannon, J. G., Turner, B., and Janal, M. (2014). Oxidative burst intensity of peripheral phagocytic cells and periodontitis in down syndrome. *J. Periodont. Res.* 49, 29–35. doi: 10.1111/jre.12075

Kim, S. C., Kim, O. S., Kim, O. J., Kim, Y. J., and Chung, H. J. (2010). Antioxidant profile of whole saliva after scaling and root planing in periodontal disease. *J. Periodont. Implant Sci.* 40, 164–171. doi: 10.5051/jpis.2010.40.4.164

Kimura, S., Yonemura, T., and Kaya, H. (1993). Increased oxidative product formation by peripheral blood polymorphonuclear leukocytes in human periodontal diseases. *J. Periodontal Res.* 28, 197–203. doi: 10.1111/j.1600-0765.1993.tb10096.x

Kinane, D. F., Preshaw, P. M., Loos, B. G., and Working Group 2 (2016). *Effects of melatonin on oxidative stress index and alveolar bone loss in chronic periodontitis. Acta Odontol. Scand.* 72, 42–47. doi: 10.1039/00016357.2013.795659

Monea, M., Mezei, T., Popsov, S., and Monea, M. (2014). Oxidative stress: a link between diabetes mellitus and periodontal disease. *Int. J. Endocrinol.* 2014:917631. doi: 10.1155/2014/917631

Muniz, F. W., Nogueira, S. B., Mendes, F. L., Rosing, C. K., Moreira, M. M., de Andrade, G. M., et al. (2015). The impact of antioxidant agents complimentary to periodontal therapy on oxidative stress and periodontal outcomes: a systematic review. *Arch. Oral Biol.* 60, 1203–1214. doi: 10.1016/j.archoralbio.2015.05.007

Myung, S. K., Ju, W., Cho, B., Oh, S. W., Park, S. M., Koo, B. K., et al. (2013). Efficacy of vitamin and antioxidant supplements in prevention of cardiovascular disease: systematic review and meta-analysis of randomised controlled trials. *BMJ* 346:f10. doi: 10.1136/bmj.f10

Nguyen, T. T., Ngo, L. C., Promsudthi, A., and Surarit, R. (2016). Salivary oxidative stress biomarkers in chronic periodontitis and acute coronary syndrome. *Clin. Oral Investig.* 21, 2345–2353. doi: 10.1007/s00784-016-2029-3

Nobili, L., Rizzo, M., Li Volli, G., D’Aiuto, F., Giglio, R. V., Barbagallo, I., et al. (2015). Lipid subclasses profiles and oxidative stress in aggressive periodontitis before and after treatment. *J. Periodont. Res.* 50, 890–896. doi: 10.1111/jre.12283

Niki, E. (2016). Oxidative stress and antioxidants: distress or eustress? *Arch. Biochem. Biophys.* 595, 19–24. doi: 10.1016/j.abb.2015.11.017

Novakovic, N., Todorovic, T., Rakic, M., Milinkovic, I., Dozic, I., Jankovic, S., et al. (2014). Salivary antioxidants as periodontal biomarkers in evaluation of tissue injury and treatment outcome. *J. Periodont. Res.* 49, 129–136. doi: 10.1111/jre.12088

Oktay, S., Chukkapalli, S. S., Rivera-Kweh, M. F., Velsko, I. M., Holliday, L. S., and Kesavalu, L. (2015). Periodontitis in rats induces systemic oxidative stress that is controlled by bone-targeted antioxidants. *J. Periodontol.* 86, 137–145. doi: 10.1002/jper.2014.140541

Onder, C., Kurgan, S., Altingöz, S. M., Başa, N., Uyanik, M., Serdar, M. A., et al. (2017). Impact of non-surgical periodontal therapy on saliva and serum levels of markers of oxidative stress. *Clin. Oral Investig.* 21, 1961–1969. doi: 10.1007/s00784-016-1984-z

Oz, H. S., and Puleo, D. A. (2011). Animal models for periodontitis. *J. Biomed. Biotechnol.* 2011:754857. doi: 10.1155/2011/754857

Özdem, M., Kiriçoğlu, F. Y., Yılmaz, H. R., Vural, H., Fentonoglu, Ö., Üz, E., et al. (2017). Antioxidant effects of melatonin in heart tissue after induction of experimental periodontitis in rats. *J. Oral Sci.* 59, 23–29. doi: 10.2334/josnd.16-0034

Panjamurthy, K., Manoharan, S., and Ramachandran, C. R. (2005). Lipid peroxidation and antioxidant status in patients with periodontitis. *Cell. Mol. Biol. Lett.* 10, 255–264.

Piskounova, E., Agathoclous, M., Murphy, M. M., Hu, Z., Huddleston, S. E., Zhao, Z., et al. (2015). Oxidative stress inhibits distant metastasis by human melanoma cells. *Nature* 527, 186–191. doi: 10.1038/nature15726

Raut, C. P., and Sethi, K. S. (2016). Comparative evaluation of co-enzyme Q10 and *Melaleuca alternifolia* as antioxidant gels in treatment of chronic periodontitis: a clinical study. *Contemp. Clin. Dent.* 7, 377–381. doi: 10.4103/0976-237X.188572

Rumbold, A. B., Crowther, C. A., Haslam, R. R., Dekker, G. A., and Robinson, J. S. (2006). Vitamins C and E and the risks of preeclampsia and perinatal complications. *N. Engl. J. Med.* 354, 1796–1806. doi: 10.1056/NEJMoa054186
Tamaki, N., Yoshino, F., Fukui, M., Hayashida, H., Yoshida, A., Kitamura, M., et al. (2015). Relationship among salivary antioxidant activity, cytokines, and periodontitis: the Nagasaki Island study. J. Clin. Periodontol. 42, 711–718. doi: 10.1111/jcpe.12438

Tartaglia, G. M., Gagliano, N., Zarbin, L., Tolomeo, G., and Sforza, C. (2017). Antioxidant capacity of human saliva and periodontal screening assessment in healthy adults. Arch. Oral Biol. 78, 34–38. doi: 10.1016/j.archoralbio.2017.02.003

Thomas, B., Madani, S. M., Prasad, B. R., and Kumari, S. (2014). Comparative evaluation of serum antioxidant levels in periodontally diseased patients: an intervention study. Contemp. Clin. Dent. 5, 340–344. doi: 10.4103/0976-237X.137938

Toker, H., Akpinar, A., Aydin, H., and Poyraz, O. (2012). Influence of smoking on interleukin-1beta level, oxidant status and antioxidant status in gingival crevicular fluid from chronic periodontitis patients before and after periodontal treatment. J. Periodontol. 83, 572–577. doi: 10.1902/jop.2011.100618

Toker, H., Ozdemir, H., Eren, K., Ozer, H., and Sahin, G. (2009). N-acetylcysteine, a thiol antioxidant, decreases alveolar bone loss in experimental periodontitis in rats. J. Periodontol. 80, 672–678. doi: 10.1902/jop.2009.080509

Tomofuji, T., Azuma, T., Kusano, H., Sanbe, T., EKuni, D., Tomaki, N., et al. (2006). Oxidative damage of periodontal tissue in the rat periodontitis model: effects of a high-cholesterol diet. FEBS Lett. 580, 3601–3604. doi: 10.1016/j.febslet.2006.05.041

Tomofuji, T., EKuni, D., Irie, K., Azuma, T., Endo, Y., Tomaki, N., et al. (2009a). Preventive effects of a cocoa-enriched diet on gingival oxidative stress in experimental periodontitis. J. Periodontol. 80, 1799–1808. doi: 10.1902/jop.2009.090270

Tomofuji, T., EKuni, D., Irie, K., Azuma, T., Tomaki, N., Maruyama, T., et al. (2011). Relationships between periodontal inflammation, lipid peroxide and oxidative damage of multiple organs in rats. Biomed. Res. 32, 343–349. doi: 10.2220/biomedres.32.343

Tomofuji, T., EKuni, D., Sanbe, T., Irie, K., Azuma, T., Maruyama, T., et al. (2009b). Effects of vitamin C intake on gingival oxidative stress in rat periodontitis. Free Radic. Biol. Med. 46, 163–168. doi: 10.1016/j.freeradbiomed.2008.09.040

Tomofuji, T., EKuni, D., Irie, K., Azuma, T., Endo, Y., Tomaki, N., et al. (2008). Oxidative damage of rat liver induced by ligature-induced periodontitis and chronic ethanol consumption. Arch. Oral Biol. 53, 1113–1118. doi: 10.1016/j.archoralbio.2008.05.015

Tonguç, M. O., Öztürk, O., Sutcu, R., Ceyhan, B. M., Kilinc, G., Sonmez, Y., et al. (2011). The impact of smoking status on antioxidant enzyme activity and malondialdehyde levels in chronic periodontitis. J. Periodontol. 82, 1320–1328. doi: 10.1902/jop.2011.100618

Torumtay, G., Kirzioglu, F. Y., Ozturk Tonguç, M., Kale, B., Calapoglu, M., and Orhan, H. (2016). Effects of periodontal treatment on inflammation and oxidative stress markers in patients with metabolic syndrome. J. Periodontol. Res. 51, 489–498. doi: 10.1111/jjrp.12328

Tothova, L., Celecova, V., and Celec, P. (2013). Salivary markers of oxidative stress and their relation to periodontal and dental status in children. Dis. Markers 34, 9–15. doi: 10.1155/2013/591765

Trivedi, S., Lal, N., Mahdi, A. A., Mittal, M., Singh, B., and Pandey, S. (2014). Evaluation of antioxidant enzymes activity and malondialdehyde levels in patients with chronic periodontitis and diabetes mellitus. J. Periodontol. 85, 713–720. doi: 10.1902/jop.2013.130066

Villa-Correia, Y. A., Isaza-Guzman, D. M., and Tobon-Arroyave, S. I. (2015). Prognostic value of 8-Hydroxy-2′-Deoxyguanosine and human neutrophil elastase/alpha1-proteinase inhibitor complex as salivary biomarkers of oxidative stress in chronic periodontitis. J. Periodontol. 86, 1260–1267. doi: 10.1902/jop.2015.150293

Vilkova, B., and Celec, P. (2009). Does Enterococcus faecalis contribute to salivary thiobarbituric acid-reacting substances? In Vivo 23, 343–345.

Watson, J. D. (2014). Type 2 diabetes as a redox disease. Lancet 383, 841–843. doi: 10.1016/S0140-6736(13)62635-3

Wei, P. F., Ho, K. Y., Hu, Y. P., Wu, Y. M., Yang, Y. H., and Tsai, C. C. (2004). The investigation of glutathione peroxidase, lactoferrin, myeloperoxidase and interleukin-1beta in gingival crevicular fluid: implications for oxidative stress in human periodontal diseases.
Weidinger, A., and Kozlov, A. V. (2015). Biological activities of reactive oxygen and nitrogen species: oxidative stress vs. signal transduction. *Biomolecules* 5, 472–484. doi: 10.3390/biom5020472

Wolfram, R. M., Budinsky, A. C., Eder, A., Presenhuber, C., Nell, A., Sperr, W., et al. (2006). Salivary isoprostanes indicate increased oxidation injury in periodontitis with additional tobacco abuse. *Biofactors* 28, 21–31. doi: 10.1002/biof.5520280103

Yang, P. S., Huang, W. C., Chen, S. Y., Chen, C. H., Lee, C. Y., Lin, C. T., et al. (2014). Scaling-stimulated salivary antioxidant changes and oral-health behavior in an evaluation of periodontal treatment outcomes. *Sci. World J.* 2014:814671. doi: 10.1155/2014/814671

Zamora-Perez, A. L., Ortiz-Garcia, Y. M., Lazalde-Ramos, B. P., Guerrero-Velasquez, C., Gomez-Meda, B. C., Ramirez-Aguilar, M. A., et al. (2015). Increased micronuclei and nuclear abnormalities in buccal mucosa and oxidative damage in saliva from patients with chronic and aggressive periodontal diseases. *J. Periodont. Res.* 50, 28–36. doi: 10.1111/jre.12175

Zare Javid, A., Seal, C. J., Heasman, P., and Moynihan, P. J. (2014). Impact of a customised dietary intervention on antioxidant status, dietary intakes and periodontal indices in patients with adult periodontitis. *J. Hum. Nutr. Diet.* 27, 523–532. doi: 10.1111/jhn.12184

Zhang, T., Andrukhov, O., Harihian, H., Muller-Kern, M., Liu, S., Liu, Z., et al. (2015). Total antioxidant capacity and total oxidant status in saliva of periodontitis patients in relation to bacterial load. *Front. Cell. Infect. Microbiol.* 5:97. doi: 10.3389/fcimb.2015.00097

**Conflict of Interest Statement:** The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

*Copyright © 2017 Tóthová and Celec. This is an open-access article distributed under the terms of the Creative Commons Attribution License (CC BY). The use, distribution or reproduction in other forums is permitted, provided the original author(s) or licensor are credited and that the original publication in this journal is cited, in accordance with accepted academic practice. No use, distribution or reproduction is permitted which does not comply with these terms.*