



Antioxidant capacity of human saliva and periodontal screening assessment in healthy adults



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ABSTRACT

Objective: Saliva plays a pivotal role as an antioxidant system, and saliva antioxidant levels are reduced in patients with periodontal disease. Recently, a biochemical test able to determine saliva antioxidant levels was proposed as predictive for oral cavity diseases, but it was not clinically tested. In this preliminary study, we evaluated the relationships between Periodontal Screening and Recordings characteristics of patients and saliva antioxidant levels measures.

Design: Thirty-nine patients (12 men, 27 women; mean age, 46 years, SD 17) attending the dental hygiene unit of a Private Clinic underwent a Periodontal Screening and Recordings examination and a saliva antioxidant levels measurement using a biochemical commercial test. The results of the clinical periodontal examination were compared to those obtained by the saliva test.

Results: Approximately 70% of patients showed a low saliva antioxidant levels value, while the other patients had Optimal/Normal values. Thirteen patients (33%) resulted positive to Periodontal Screening and Recordings test. Using Periodontal Screening and Recordings values as gold standard, the saliva antioxidant levels test correctly classified 52.6% of patients; sensitivity was 84.6%, specificity was 36%. **Conclusions:** The saliva antioxidant levels test had a good sensitivity when compared to the gold standard; this finding corroborates the hypothesis that alterations of the oral antioxidant levels are related to periodontal disease. The reduced specificity shows that saliva antioxidant levels test could detect alterations predisposing to periodontal disease before clinically evident aspects.

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1. Introduction

Periodontal disease (PD), outlined as “An infectious disease resulting in inflammation within the supporting tissues of the teeth, progressive attachment and bone loss” from the [American Academy of Periodontology Task Force \(2015\)](#), is one of the most prevalent bacterial-induced chronic diseases affecting the majority of adults ([Dye, 2012](#); [Zare Javid, Seal, Heasman, & Moynihan, 2014](#)). One of its forms, severe periodontitis, with dental attachment loss and periodontal depth larger than 6 mm, was the sixth-most prevalent disease condition, affecting 10.8% or 743 million people worldwide ([Kassebaum et al., 2014](#)).

Consequence of untreated PD is tooth loss. The patient-reported outcome measures of tooth loss are reduced functional capacity (e.g. chewing or biting), reduced self-esteem and social relationships, thus resulting in reduced people life quality ([Petersen & Ogawa, 2012](#)). Moreover, inflammatory periodontal diseases may be a risk factor for different multiple systemic conditions like metabolic syndrome-MetS ([Torumtay et al., 2016](#); [Watanabe & Cho, 2014](#)) and various chronic diseases ([Otomo-Corgel, Pucher, Rethman, & Reynolds, 2012](#)). Interestingly, evidence that mobile oral microbiome should be a reservoir for extra-oral infections and systemic dissemination of pathogenic toxins is increasing, although studies linking oral bacteria to extra-oral infections are still at the stage of association ([Han & Wang, 2013](#)). At the same time, recent genome-wide association studies found some suggestive genetic polymorphisms associated with specific phenotypes of chronic periodontitis, thus posing the bases for future

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investigations about the pathways leading to this disease (Shaffer et al., 2014).

In terms of clinical recommendations, maintenance of good oral hygiene, especially for immune-compromised patients, is crucial for controlling total bacterial load to prevent bacterial dissemination. Progression of periodontal disease is dependent on the host immune response and individual susceptibility (Kornman, 2008; Muniz et al., 2015). Oxidative stress (OS), due to imbalance between oxidants load and antioxidant capacity, is a potential mediator of the progression of different seemingly unrelated diseases including cancer, Parkinson's and Alzheimer's diseases, atherosclerosis, myocardial infarction as well as periodontal disease (Avezov, Reznick, & Aizenbud, 2015; Torumtay et al., 2016).

OS is one of the most important causative factors for the induction of cell apoptosis, bringing the cells into a state similar to senescence called stress-induced premature senescence. In vitro experimental studies show that OS is possibly correlated with the development of periodontal diseases (Bañasová et al., 2015; Kiyoshima et al., 2012). It was demonstrated that OS affects gingival fibroblasts by inhibiting cell viability and proliferation, and affecting cell morphology, therefore influencing their ability for extracellular matrix remodeling in the periodontal tissues (Colombo et al., 2012).

In the inflammatory process generated from the microbial plaque that accumulates around the gingival margin, polymorphonuclear leucocytes are activated, and reactive oxygen species (ROS) and reactive nitrogen species (RNS) are produced as a result of bacterial destruction. In predisposed persons that have an aberrantly exaggerated inflammatory/immune response, the production of ROS and RNS can result in inadvertent or collateral host tissue damage (Zare Javid et al., 2014).

Not only OS but also the redox balance may be relevant in pathological conditions. Decreased antioxidant ability might render the cells more vulnerable to OS in the oral cavity and might impair the reparative and regenerative potential of gingival tissues (Bergstrom, 2004; Cattaneo et al., 2000; Poggi, Rota, & Boratto, 2002).

Saliva plays a pivotal role as an antioxidant system, including various molecules and enzymes, of which the most important are the uric acid molecule and the peroxidase enzyme (Nagler, Klein, Zarzhevsky, Drigues, & Reznick, 2002). Decreased salivary antioxidant ability to reduce OS was related to alterations of the oral mucosa, including oral cancer (Dayan, Hirshberg, Kaplan, Rotem, & Bodner, 1997; Giebultowicz, Wroczynski, & Samolczyk-Wanyura, 2011; Nishioka, Nishi, & Kyokane, 1981) and lichen planus (Batu et al., 2015; Shirzad et al., 2014).

Saliva antioxidant levels (SAL) are significantly lower in chronic periodontitis patients when compared to periodontally healthy individuals (Bañasová et al., 2015), and it was demonstrated that PD is associated with reduced salivary antioxidant status and increased oxidative damage in the oral cavity (Sculley & Langley-Evans, 2003). Therefore, SAL and PD seem to be associated with one another, leading to increased oxidative damages in the oral environment (Brock, Butterworth, Matthews, & Chapple, 2004).

Interestingly, the composition of saliva varies in different local and systemic diseases, and salivary antioxidant potential may reflect many pathophysiological states (Eisenberg et al., 2008; Nagler et al., 2002; Reznick, Hershkovich, & Nagler, 2004; Zloczower, Reznick, Zouby, & Nagler, 2007). The removal and control of ROS and RNS is important in preventing destruction of periodontal tissue (Chapple et al., 2002), and it is currently successfully treated by removing the supra- and subgingival biofilm by scaling and root planing combined with adequate periodontal support maintenance (Ramfjord, Knowles, Nissle, Burgett, & Shick, 1975; Torumtay et al., 2016). The early identification of predisposed patients able to develop PD and/or

its severe form is very interesting in both clinical and socio-economical terms for researcher and dental professionals. Thus, on the wave of recent research suggestions on molecular aspects of pathogenesis of periodontitis (Meyle & Chapple, 2015), it is important, first of all, to understand if SAL measures could be representative of patient incipient dysbiosis.

Benedetti, Primiterra, Finco, Canestrari, and Cornelli (2014) recently validated a biochemical test able to determine SAL in a simple, fast and economical way, and proposed its use as a predictive test for both local oral cavity and general body diseases. To the best of our knowledge, no clinical investigation compared the actual results of SAL assessments to some recognized gold standard for PD. Therefore, the aim of the present preliminary investigation is to evaluate a clinical gold standard assessment for PD, Periodontal Screening and Recordings, (PSR); in comparison to SAL measures in terms of detection of periodontal problems. The vast majority of academicians and professionals consider PSR beneficial in terms of disease detection, record keeping, cost effectiveness and patient education (Landry & Jean, 2002). Furthermore, PSR is a dental community appreciated index (Primal, Esther, & Boehm, 2014).

In the present study, the PSR characteristics of a group of adult patients attending a private practice dental clinic were analyzed and compared to SAL measures taken in the same occasion (Benedetti et al., 2014). The use of quantitative clinical evaluations can reduce inter-operator variability, accelerate the diagnosis of PD, and promote patient's motivation (Wang, Schipper, Velly, Mohit, & Gornitsky, 2015).

2. Material and methods

2.1. Patient selection

From the beginning to the end of September 2015, 39 Caucasian patients (12 men and 27 women; mean age, 46 years, SD 16.9 years; age range, 18 to 80 years) were selected from patients attending the dental hygiene unit of a Private Clinic in Segrate, Milan (Italy). The patients underwent a Periodontal Screening and Recordings examination (PSR) and a saliva sample collection to perform a SAL measurement (Benedetti et al., 2014). The procedures were performed by two dental hygienists and a dentist; each professional made only one of the two examinations on each patient, and was blind to the results of the other test. Patients were randomly allocated to the professionals.

Overall, the Study Inclusion Criteria were to enroll patients of both genders, older than 18 years, with a moderate-good oral health condition, encountering a combination of the following points:

1. With 28 teeth at least
2. DMFT index ≤ 50
3. Tooth mobility Modified Miller index ≤ 1 (Wasserman, Geiger, & Turgeon, 1973)
4. Plaque index $\leq 25\%$ (O'Leary, Drake, & Naylor, 1972)
5. Absence of active caries (Gomez, Tellez, Pretty, Ellwood, & Ismail, 2013)
6. Motivation to maintenance program
7. No dental treatment during the preceding two months period.

The patients were asked to update their personal and medical history in particular about alcohol and tobacco use; "drinkers" were all patients who declared to had been consuming more than half a liter/day of generic alcoholic beverages continuously for at least one year, while "smokers" were all patients who declared to be habitual tobacco users. These last ones were excluded. All subjects signed a written consent form given thorough explanation

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