The Association of Clinical and Microbiologic Parameters with Histologic Observations in Relatively Healthy Peri-Implant Conditions—A Preliminary Short-Term In Vivo Study

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Purpose: To determine whether clinical findings—bleeding on probing, pocket depth, recession, and bacterial sampling—correlate with histologic outcomes in relatively healthy peri-implant soft tissues in people. ***Materials and Methods:*** In this cross-sectional study, a convenience sample of 20 edentulous subjects received two endosseous mandibular implants each. The abutments were either zirconia (ZrO_2) or titanium (Ti) (nonsubmerged implant placement, within-subject comparison, left-right randomization). Sulcular bacterial sampling and assessment of probing pocket depth, recession, and bleeding on probing were performed 3 months postsurgery. Mucosal biopsy specimens were obtained, and the blood vessel density and a score on an inflammation grading scale were determined. ***Results:*** Simple linear and linear regression models revealed that the clinical or microbiologic parameters were not associated with either of the histologic parameters. The soft tissues impressed as healthy, regardless of the abutment material. ***Conclusions:*** The peri-implant mucosa around ZrO_2 and Ti abutments was considered healthy in most situations when examined histologically after 3 months but showed variation in clinical and microbiologic parameters. *Int J Prosthodont 2014;27:573–576. doi: 10.11607/ijp.3922*

From a limited number of clinical studies, zirconia (ZrO_2) and titanium (Ti) abutments seem to elicit a similar soft tissue response,[1–3] but studies involving human histology are scarce. This study focused on the response and health of the soft tissues toward ZrO_2 and Ti implant abutments in people as observed clinically, microbiologically, and histologically. Possible associations between outcome parameters were the primary subject of interest in order to elucidate whether they can reliably reflect peri-implant soft tissue health.

Materials and Methods

Twenty edentulous implant-overdenture candidates, 9 men and 11 women, between 39 and 76 years of age (mean: 56.4 years), received two mandibular implants. The inclusion criteria, surgical and restorative procedures, and materials have been previously described.[4] The implants were immediately provided with one ZrO_2 (experimental) and one Ti abutment that functioned as healing abutments (nonsubmerged implant placement, within-subject comparison, left-right randomization, allocation revealed directly after implant placement; Fig 1). Clinical, microbiologic, and histologic data were collected after 3 months of permucosal healing. Two days preoperatively and 2 weeks postoperatively, patients rinsed with 0.2% chlorhexidine solution. Two weeks after surgery, brushing was allowed but no professional cleaning was performed during the experimental period, and the old prostheses were adapted to fit over the abutments.

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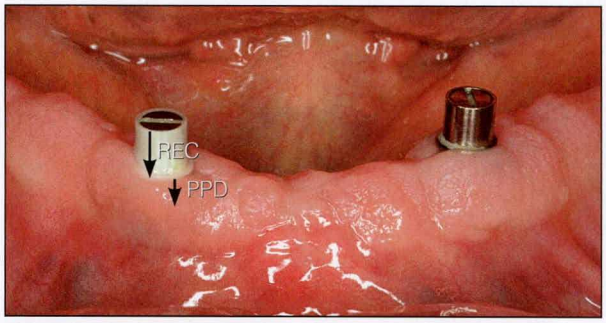


Fig 1 Zirconia and titanium experimental abutments in situ after 3 months. Recession (REC) is measured from the edge of the implant to the mucosal margin. The pocket probing depth (PPD) is measured from the mucosal margin to the clinical pocket.

Table 1 Quantitative Histologic Scoring System for Inflammation

Score	Description
1	Masses of inflammatory cells
2	Many inflammatory cells, showing some fibroblasts
3	Immature connective tissue, showing fibroblasts with few inflammatory cells
4	Normal appearance of connective tissue with few inflammatory cells

Clinical and Microbiologic Parameters

Probing pocket depth (PPD), recession (REC), and bleeding on probing (BOP) were assessed at two sites per implant (midbuccal and mesial). A plastic periodontal probe with 0.25 N of calibrated probing force was used (Click-probe, KerrHawe). BOP is presented as the percentage of implants that demonstrated either midbuccal or mesial BOP. Sulcular plaque samples were obtained by performing a circumferential motion (360 degrees) in the peri-implant sulci with a sterilized single-use plastic scaler (Implacare, Hu-Friedy).

Detection and counting of seven periodontal pathogenic microorganisms were performed using real-time polymerase chain reaction (PCR). The total bacterial load was considered to be the microbiologic outcome measure.

Histologic Parameters

Mesial and distal triangular soft tissues from 17 patients could be retrieved using a scalpel (68 specimens). They were embedded in Paraplast paraffin (Klinipath). Sections of 6 μm were cut and stained with hematoxylin-eosin (HE). To elucidate the presence of blood vessels, collagen intravenous staining of their vascular basement membrane was performed.

Regions of interest (ROI) measuring 200 × 200 μm at the soft tissue–abutment interface were defined for two sections per abutment: a mesial and a distal one. The number of blood vessels within each ROI was counted to determine vascular density. Mesial and distal sites were averaged. Inflammation was also scored on a 4-point scale (Table 1). A higher score represents a better (ie, reduced inflammatory) response.

Results

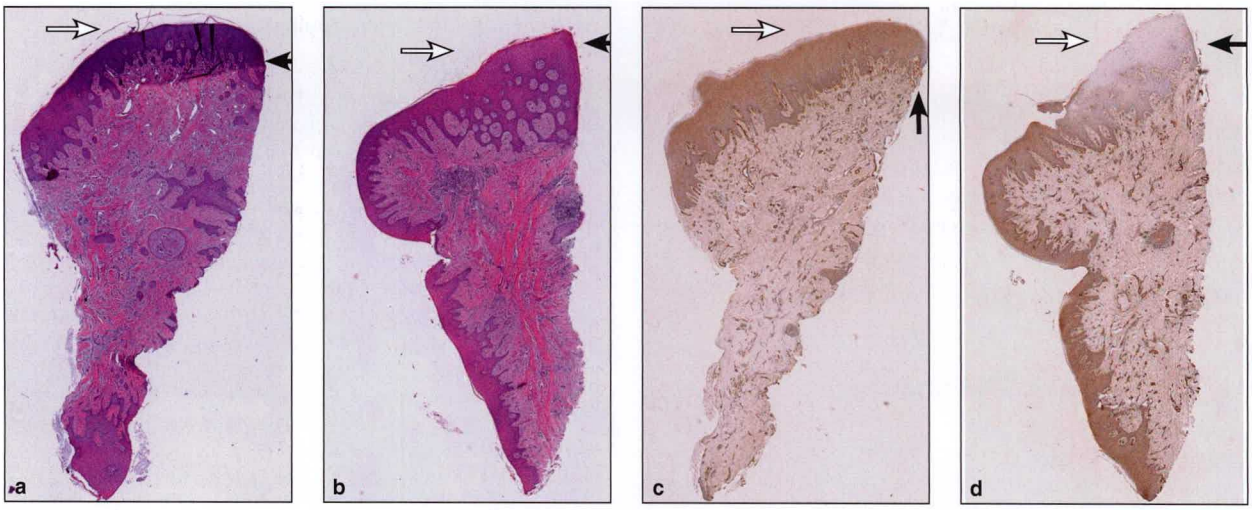
After histologic preparation, paired samples from 14 patients were deemed suitable for further analysis. Clinical and microbiologic data at 3 months in one participant could not be recorded because of a breach of protocol. Consequently, differences between observations around the two abutment types and associations between parameters could be calculated in 13 patients (Fig 2).

Mean scores are presented in Table 2. No statistically significant differences between abutment types were observed for any parameter. Histology generally revealed healthy soft tissues. Pearson correlation coefficients between all parameters are presented in Table 3. The correlations between the clinical and microbiologic parameters and the two histologic parameters are rather low and not statistically significant, with the exception of the association between REC and PPD for Ti and ZrO_2 abutments.

The four linear regression models explained between 24.9% and 65.5% of the observed variation, and none of the clinical or microbiologic parameters were statistical significantly associated with either the blood vessel density or the inflammation grading score.

Discussion

No differences in clinical, microbiologic, and histologic parameters between zirconia and titanium abutments were observed. No obvious associations between clinical and microbiologic findings on the one hand and histologic findings on the other hand were seen. This raises questions toward the appropriateness of periodontal parameters to reflect peri-implant soft tissue health. They are not always in sync.[5]



Figs 2a to 2d Representative images from the study. **(a)** Hematoxylin-eosin (HE) staining, showing the intact intraoral mucosa *(white arrow)* and the barrier epithelium *(black arrow)* bordering the titanium abutment. **(b)** HE staining, showing the intact intraoral mucosa *(white arrow)* and the barrier epithelium *(black arrow)* bordering the zirconia abutment. **(c)** Collagen intravenous (IV) staining *(brown)*, showing the intact intraoral mucosa *(white arrow)* and the barrier epithelium *(black arrow)* bordering the titanium abutment. **(d)** Collagen IV staining *(brown)*, showing the intact intraoral mucosa *(white arrow)* and the barrier epithelium *(black arrow)* bordering the zirconia abutment.

Table 2 Mean Values (SDs) for Clinical, Microbiologic, and Histologic Parameters (n = 13 Patients and 26 Abutments)*

	Zirconia abutment	Titanium abutment	*t*	*P* value
Blood vessel density	20.9 (4.4)	21.1 (3.2)	–0.27	.79
Histology grading scale	3.1 (0.6)	3.2 (0.6)	–0.62	.55
Bacterial load	2.0×10^6 (5.5×10^6)	9.2×10^6 (26.4×10^6)	0.98	.35
Mean probing pocket depth	1.8 (0.5)	2.1 (0.8)	–1.39	.19
Mean recession	2.7 (0.5)	2.7 (1.1)	0.0	> .99
Mean bleeding on probing	38.5% (36.3%)	62.5% (46.3%)	–1.20	.25

*Paired samples *t* test.

Table 3 Matrix of Pearson Correlations Between Clinical, Microbiologic, and Histologic Parameters (n = 13 Patients and 26 Abutments)

	HGS	BL	mPPD	mREC	mBOP*
Blood vessel density (BVD)					
ZrO_2	r = -0.14, P = .65	r = 0.38, P = .20	r = 0.10, P = .74	r = -0.35, P = .24	r = 0.26, P = .39
Ti	r = 0.13, P = .68	r = -0.19, P = .54	r = 0.06, P = .86	r = 0.01, P = .96	r = 0.06, P = .84
Histology grading scale (HGS)					
ZrO_2		r = -0.20, P = .96	r = -0.19, P = .53	r = 0.42, P = .15	r = 0.22, P = .47
Ti		r = 0.46, P = .11	r = 0.30, P = .32	r = -0.32, P = .29	r = -0.10, P = .74
Bacterial load (BL)					
ZrO_2			r = -0.12, P = .63	r = -0.01, P = .98	r = -0.10, P = .68
Ti			r = 0.24, P = .33	r = 0.05, P = .84	r = -0.36, P = .14
Mean probing pocket depth (mPPD)					
ZrO_2				r = -0.54, P = .02**	r = 0.10, P = .66
Ti				r = -0.62, P = .01**	r = -0.22, P = .37
Mean recession (mREC)					
ZrO_2					r = 0.20, P = .41
Ti					r = -0.22, P = .37

*mBOP = mean Bleeding on Probing.
** = statistically significant.

Conclusions

It was concluded that zirconia and titanium implant abutments elicit a similar soft tissue response when judged clinically, microbiologically, and histologically in people, but the parameters do not correlate. This gave rise to concerns as to the sensitivity and specificity of clinical and microbiologic parameters as indicators of the peri-implant soft tissue status in relatively healthy conditions in vivo.

Acknowledgments

The authors are grateful to Prof Dr Edwin van den Heuvel of the Department of Medical Statistics of the University Medical Center Groningen for providing statistical support.

This research was made possible by a grant from Astra Tech AB (Mölndal, Sweden), which also provided the experimental abutments without cost, and by the support of the authors' institutions. The authors reported no conflicts of interest related to this study.

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Literature Abstract

Alzheimer's disease and periodontitis—An elusive link

Alzheimer's disease (AD) is the most prevalent form of dementia, which poses a health problem worldwide. However, the etiology and pathophysiology of this complex neurodegenerative disorder has not been identified. The objective of this review thus aimed to clarify the possible role of periodontitis in exacerbating AD. A PubMed search incorporating relevant systematic reviews, meta-analyses, and original articles in English from 1994 to 2012 was used. Both human and animal studies were considered. A distinct hallmark of AD is the formation of extracellular amyloid β-peptide (AβP) plaques from the proteolytic cleavage of the amyloid precursor protein (APP) and intraneuronal neurofibrillary tangles (NFTs) of hyper-phosphorylated tau protein. The interplay of the above encourages release of pro-inflammatory mediators within the cerebral microvasculature leading to impaired degradation and clearance and subsequent loss of synaptic dysfunction and neurodegeneration. Neural damage can be induced via the exaggerated inflammation found in AD. Periodontitis, even though primarily a local disease, is able to sustain a low-grade systemic inflammation through the release of pro-inflammatory cytokines. Two plausible links are found between AD and periodontitis through periodontitis preceding systemic inflammation or infection, as well as a bacterial and viral influence. This systemic infection or inflammation is postulated to bypass a compromised blood-brain barrier and via peripheral nerves leading to microglial activation, tau protein phosphorylation, increased APP and AβP, alongside platelet aggregation and atherogenesis. This contributes to oxidative damage and inflammation, which are important features of AD. The author proposed that inflammation may be the central operating mechanism and considers that periodontitis may be a potential risk factor for the development of AD. Given the limitations of evidence available, the question of causality remained unanswered and could be clarified only by further research.

Gurav AN. *Rev Assoc Med Bras* 2014;60:173–180. **References:** 73. **Reprints:** Department of Periodontics Tatyasaheb Kore Dental College & Research Centre New Pargaon Kolhapur - 416137 Maharashtra State India. Fax: +91 230 2477082. Email: dr_abhijitg@yahoo.co.in
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