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# Reduction of Bacterial Proliferation by Zirconium Collar in Dental Implants

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#### Authors' contributions

This work was carried out in collaboration between all authors. Author AM designed the study and the protocol, and performed the surgery. Authors FA and SB managed the analyses of the study and revised the manuscript. Author SZ performed the statistical analysis and managed the literature searches. Author AO designed the study, co-performed the surgery, wrote the protocol and the first draft of the manuscript and revised the manuscript. All authors read and approved the final manuscript.

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## **ABSTRACT**

The peri-implant bacterial colonization is one of the current major problems facing dental implants with no actual protocols for prevention. The use of zirconium for bacterial eradication has already been reported and discussed in the dental literature. In this study we evaluated for the first time the ability of a "hybrid" dental implant model – where the implant collar is made out of titanium and zirconium - to reduce the peri-implant bacterial colonization, using traditional implants from the same manufacturer as controls. The results of microbiological analysis and the evaluation of the classic parameters of an implant success confirmed that, in all the 30 patients in this study, the zirconium collar had a vital role in reducing peri-implant bacterial colonization, and that the "hybrid"

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implants show lower plaque index values, less bleeding and less marginal bone loss than the traditional implants. Our data therefore suggest that a zirconium collar can effectively reduce the bacterial colonization around a titanium implant favoring a better long-term prognosis.

Keywords: Dental implants; Zirconium collar; peri-implant bacterial count.

### 1. INTRODUCTION

In the literature there are several studies highlighting the problem of peri-implant bacterial colonization, especially those affecting the transmucosal collar [1-4]. The myriad of choices from implants with different profiles and the use of "bone-level" versus subcortical insertion regulates the emergence profile and the dimensional stability of the peri-implant transmucosal tunnel. To date, there are no protocols to predetermine the possible periimplant bacterial proliferation according to the type of prosthesis or of choice on the emergence profile [5]. In this same research direction, the use of zirconium (Zr) as an alternative to traditional metal-ceramic prostheses in general dentistry has already shown benefits in the control of tissue stability in the medium to long term [6]. Subsequently, its use in implant prosthetics increased its long-term stability together with the proliferation of the surrounding tissues [7]. Implants entirely in Zr have been used and studied since 1975 and regularly researched since the mid-1980s [8,9]. However, their structural and mechanical capacities and their lower fracture resistance compared to titanium (Ti) allowed their limited use only in cases where occlusal forces were compatible with this type of fixture [10,11].

The subsequent introduction of "hybrid "implants, i.e. a Ti structure and an intimately adherent Zr collar, showed broad advantages in fields of dental implants from anterior esthetic segments to the more complex posterior restorations even when bruxism or parafunctional habits are involved. In addition, it was shown that implants with Zr collar promote the proliferation and adhesion of osteoblasts and fibroblasts [12].

Of note, all issues related to the structural fragility of the Zr do not apply in this "hybrid" type of implants since the close adherence of the Zr and Ti metals form a single body [13] which prevents warping, twisting or compression that may lead to fracture unless the problems are related to the fractures of the actual implant neck as mentioned in the literature [14,15].

In this study, we tested the behavior of two types implant from the point of view of the more classical mechanical and biological parameters, in particular related to the expression of the bacterial load in the implant sites. Our first "test" group consisted of implants in Ti + Zr collar while the second "control" group constituted of conventional Ti implant from the same manufacturer.

#### 2. MATERIALS AND METHODS

In this study 30 patients were involved over the past 24 months under our observation at the clinic of the Department of Surgical Sciences and Integrated Diagnostic (Odontostomatology unit) University of Genoa, after obtaining their informed consent (clinical trial certificate number: HSM-16-H5462746). Patients were selected according to their need of a partial arch rehabilitation - maxillary or mandibular. In order for each individual to receive a test and a control implant, at least two single posterior teeth premolars or molars - were required to replace. A total of 60 implants were used, TBR® (Toulouse, France), 2 for each patient, 30 implants "Z1 infinity®", with zirconium collar 1.5mm, as " test" (A) , and another 30 implants "infinity"® traditional completely titanium (B), as control (Fig. 1 : systems "A" & "B"). Both implants had a 3.5 diameter and 10-11.5-13 mm length.



Fig. 1. Implant Zn+Ti, type A. Implant Ti, type B

The selection of patients included 13 males aged between 36 and 65 years (mean age 49) and 17 females aged between 38 and 69 years (mean age 51). We have excluded from the study patients who had denied consent, patients with poor oral hygiene having full mouth Plaque Score (FMPS)  $\geq$  20 % before surgery, patients with compromised periodontium or local soft tissue infections, smokers (even occasional), patients with psychiatric illnesses and pregnant women. Fig. 2 shows a selected pre-operative panoramic radiograph from a 54 year-old patient.

According to dental implants classic technique [16], after nerve block local anesthesia with 2% Mepivacaine, a full-thickness flap was reflected and the implants "A" and "B" were inserted with

shoulder protocol in the edentulous crests with bone-level seating (Ti implants "B") or 0.5 mm sinking below the cortical bone level (Ti+Zr implants "A") (Fig. 3: Post-operative panoramic Horizontal mattress suturing radiograph). technique (Vycril®) was used to suture the implant site. The implants were loaded three months post insertion. All patients were subjected to periodic inspections at 30, 60 and days after surgery with panoramic radiographs all with the same X-ray machine. (Fig. 4: 3 months post-loading panoramic radiograph). The biological parameters evaluated at 3 months post-loading included implant mobility (yes/no), Plaque Index (PI) [17], Bleeding Index (BI), and marginal bone loss (MBL).



Fig. 2. A pre-operative panoramic radiograph from a 54 year-old patient



Fig. 3. A post-operative panoramic radiograph



Fig. 4. A 3 months post-loading panoramic radiograph

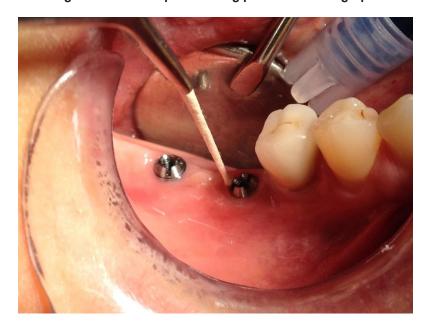


Fig. 5. Bacterial collection from the peri-implant sulcus by paper points

At the same time the peri-implant tissues were evaluated for biological parameters. The implant site was isolated with cotton rolls and a sterile paper point was inserted in the gingival sulcus and removed after 10 seconds (Fig. 5). Following the withdrawal, the paper points (2 for each patient, one for "A" and one for "B") were placed in a sterile tube and sent to the microbiology laboratory of the department for bacterial flora analysis. The concentrations of the following microorganisms were evaluated as described elsewhere [18-19]: *Porphyromonas gingivalis* (Pg), *Tannerella forsythia* (Tf),

Prevotella intermedia (Pi), Aggregatibacter actinomycetemcomitans (Aa).

## 2.1 Statistical Analysis

Data from bacterial load, plaque index, bleeding index and marginal bone loss was analyzed by two-way ANOVA test utilizing Minitab software. Mann-Whitney U-test analyses were used to detect on which of the outcome measures the groups differed. Data are shown as average  $\pm$  standard deviation. The criterion for statistical significance in all tests was p  $\leq$  0.05.

Table 1. Bacterial load with zirconium (A), and traditional titanium (B) implants

Microrganisms at 6 months	Median Bacterial load implants "A" (GE/ml)	Median Bacterial load implants "B" (GE/ml)	Statistical Mann- Whitney U-test p value
T. forsythia	8.6x10 <sup>3</sup>	2.7x10⁴	0.025
P. gingivalis	$3.8x10^2$	1.1x10 <sup>3</sup>	0.291
P. intermedia	4.0x10 <sup>3</sup>	6.1x10 <sup>3</sup>	0.148
A. actinomycetemcomitans	1.4x10 <sup>3</sup>	1.7x10 <sup>3</sup>	0.096

#### 3. RESULTS

The laboratory tests results of the bacterial load at 6 months post-implantation for "A" and "B" implants in individual patients have shown that, for all tested microorganisms and for all of the 30 patients, there have been lower level of colonization around implants "A" (Table 1). Zirconia implants showed significantly lower colonization from T. forsythia compared to titanium implants (8.6x10<sup>3</sup> and 2.7x10<sup>4</sup> respectively; P=0.025) and less significance with other bacterial species. The periodic inspections showed better tissue dimensional stability around Zirconia implants compared to titanium implants in all 30 patients: reduced of bacterial plaque  $(0.37 \pm 0.33 \text{ vs. } 0.49 \pm 0.31)$ , less marginal bone loss (1.05  $\pm$  0.89 vs. 1.21  $\pm$  0.99) and less bleeding (0.18  $\pm$  0.22 vs. 0.29  $\pm$  0.23), however results did not show significance (P=0.093, 0.342 and 0.053 respectively: Tables 2 and 3). Moreover, the survey also showed a closer tissue adherence to the zirconium collar, and in 11 cases we found an increase in connective tissue that completely covered the healing screw and in 5 cases we noticed well vascularized D3 type bone with an increase in peri-implant bone peaks visible radiographically.

Table 2. Plaque and bleeding indices with zirconium (A), and traditional titanium (B) implants

6 months Follow-up	Plaque Index	Bleeding Index
Implants "A"	$0.37 \pm 0.33$	0.18 ± 0.22
Implants "B"	$0.49 \pm 0.31$	$0.29 \pm 0.23$
Significance	0.093	0.053

Table 3. Marginal bone loss with zirconium (A), and traditional titanium (B) implants

6 months Follow-up	Marginal bone loss
Implants "A"	1.05 ± 0.89
Implants "B"	1.21 ± 0.99
Significance	0.342

#### 4. DISCUSSION AND CONCLUSION

The use of zirconium for the bacterial eradication in dentistry has already been reported and discussed in the literature [20-24]. In this study the authors propose to use the zirconium material in dental implantology, through the employment of "hybrid" implants (titanium screw with zirconium collar). That latter, when compared to conventional Titanium implants, have proven for the first time very effective in reducing bacterial colonization around the dental implants, in all of our 30 randomly selected patients.

According to our experimental findings, we can define hybrid implant an excellent substitute of conventional implants in pure titanium, since these former appear on one hand to ensure the physical properties of titanium and on other the cosmetics and antibacterial properties of the zirconium. Moreover, the use of this hybrid type of implants ensures, if properly positioned, a connective tissue seal and a structural integrity with the prosthetic device (Fig. 6). However, in cases where the marginal gingiva compromised we recommend its avoidance due to the shiny titanium collar. Our results confirm the performance of a Zirconium collar implant that literature report to show biocompatibility [25] and connective tissue adhesion similar to that seen on the machined titanium surface, however with limited plague formation and of course better esthetics [26].

In addition, when a traditional prosthesis in zirconium on natural elements is used, the collar would reduce the bacterial colonization and the development of the biofilm. Besides, antioxidants would improve mitochondrial functionality and regulate apoptosis [27].

On the other hand, inflammation always occur due to trauma post implant insertion besides the oxidative stress in the immunological response to bacterial infection. For such situation, literature has consistently recommended the use of antioxidant treatment to prevent or decrease the onset of oxidative stress [28].



Fig. 6. Gingival seal around the implants

Since the most common causes of implant failure is peri-implantitis caused by bacterial biofilm colonization on the surfaces of dental implants. research have been hunting for the surface that causes the least bacterial adhesion on an osteointegrated titanium alloy implant. Zirconium such as in our and comparable studies [9,21], have been demonstrated to reduce adhesion of certain causative bacterial species. Silver hydroxyapatite nanocoatings were found to have antibiofilm properties while conserving the hydroxyapatite biocompatibility [29]. Anastase nanocoating have also been found to have low colonization potential in addition to its desirable genetic effects on osteoblasts [30,31]. Despite the promise of each type of coating and the technology behind it, a wide comprehensive study including all types of antibacterial coatings is still missing for conclusive recommendations.

In conclusion, our study demonstrated and discussed solid data confirming the superiority of a dental implant with a hybrid zirconium collar, yet we recommend further investigations as well as a longer term follow—up.

#### **COMPETING INTERESTS**

Authors have declared that no competing interests exist.

#### **REFERENCES**

- Tamura N, Ochi M, Miyakawa H, Nakazawa F. Analysis of bacterial flora associated with peri-implantitis using obligate anaerobic culture technique and 16S rDNA gene sequence. Int J Oral Maxillofac Implants. 2013;28(6):1521-9. DOI: 10.11607/jomi.2570
- Dabdoub SM, Tsigarida AA, Kumar PS. Patient-specific analysis of peri-implant microbiomes. J Dent Res. 2013;92 (12 Suppl):168S-175S. DOI: 10.1177/0022034513504950
- 3. Zheng H, Xu L, Wang Z, Li L, Zhang J, Zhang Q, Chen T, Lin J, Chen F. Subgingival microbiome in patients with healthy and ailing dental implants. Sci Rep. 2015;16:10948.

DOI: 10.1038/srep10948

- Crippa R, Paglia M, Ferrante F, Ottonello A, Angiero F. Tongue-tie assessment: Clinical aspects and a new diode laser technique for its management. Eur J Paediatr Dent. 2016;17(3):220-222.
- Wang HL, Garaicoa-Pazmino C, Collins A, Ong HS, Chudri R, Giannobile WV. Protein biomarkers and microbial profiles in periimplantitis. Clin Oral Implants Res. 2016;27(9):1129-36. DOI: 10.1111/clr.12708
- Gahlert M, Röhling S, Wieland M, Eichhorn S, Küchenhoff H, Kniha H. A comparison study of the osseointegration of zirconia and titanium dental implants. A biomechanical evaluation in the maxilla of pigs. Clin Implant Dent Relat Res. 2010;12(4):297-305.
   DOI: 10.1111/j.1708-8208.2009.00168.x
- Bormann KH, Gellrich NC, Kniha H, Dard M, Wieland M, Gahlert M. Biomechanical evaluation of a microstructured zirconia implant by a removal torque comparison with a standard Ti-SLA implant. Clin Oral Implants Res. 2012;23(10):1210-6. DOI: 10.1111/j.1600-0501.2011.02291.x
- Cranin AN, Schnitman PA, Rabkin SM, Onesto EJ. Alumina and zirconia coated vitallium oral endosteal implants in beagles. J Biomed Mater Res. 1975;9(4): 257-62.
   DOI: 10.1002/ibm.820090429
- Albrektsson T, Hansson HA, Ivarsson B. Interface analysis of titanium and zirconium bone implants. Biomaterials.

1985;6(2):97-101.

- Wenz HJ, Bartsch J, Wolfart S, Kern M. Osseointegration and clinical success of zirconia dental implants: A systematic review. Int J Prosthodont. 2008;21(1):27-
- Apratim A, Eachempati P, Krishnappa Salian KK, Singh V, Chhabra S, Shah S. Zirconia in dental implantology: A review. J Int Soc Prev Community Dent. 2015;5(3): 147-56.
  - DOI: 10.4103/2231-0762.158014
- 12. Bianchi AE, Bosetti M, Dolci G Jr, Sberna MT, Sanfilippo S, Cannas M. In vitro and in vivo follow-up of titanium transmucosal implants with a zirconia collar. J Appl Biomater Biomech. 2004;2(3):143-50.
- Tetè S, Mastrangelo F, Bianchi A, Zizzari V, Scarano A. Collagen fiber orientation around machined titanium and zirconia dental implant necks: An animal study. Int

- J Oral Maxillofac Implants. 2009;24(1):52-8.
- Alqahtani F, Flinton R. Postfatigue fracture resistance of modified prefabricated zirconia implant abutments. J Prosthet Dent. 2014;112(2):299-305.
   DOI: 10.1016/j.prosdent.2013.08.023
- Protopapadaki M, Monaco EA Jr, Kim HI, Davis EL. Comparison of fracture resistance of pressable metal ceramic custom implant abutment with a commercially fabricated CAD/CAM zirconia implant abutment. J Prosthet Dent. 2013;110(5):389-96. DOI: 10.1016/S0022-3913(09)60043-3
- 16. Deppe H, Wolff C, Bauer F, Ruthenberg R, Sculean A, Mücke T. Dental implant surfaces after insertion in bone: An *in vitro* study in four commercial implant systems. Clin Oral Investig. Oct 24; 2017. DOI: 10.1007/s00784-017-2262-4
- 17. O'Leary TJ, Drake RB, Naylor JE. The plaque control record. Journal of Periodontology. 1972;43(1):38.
- Savarino E, Ottonello A, Tolone S, Bartolo O, Baeg MK, Farjah F, Kuribayashi S, Shetler KP, Lottrup C, Stein E. Novel insights into diagnostic procedures. Ann N Y Acad Sci. 2016;1380(1):162-177.
   DOI: 10.1111/nvas.13255
- Johansson K, Jimbo R, Östlund P, Tranæus S, Becktor JP. Effects of Bacterial contamination on dental implants during surgery. Implant Dent. 2017;26(5):778-789.
   DOI: 10.1097/ID.00000000000000660
- Zhou J, Pavon-Djavid G, Anagnostou F, Migonney V. Inhibition of *Porphyromonas* gingivalis adherence on titanium surface grafted by poly (sodium styrene sulfonate). ITBM-RBM. 2007;28:42-48.
- Scarano A, Piattelli M, Caputi S, Favero GA, Piattelli A. Bacterial adhesion on commercially pure titanium and zirconium oxide disks: An *in vivo* human study. J Periodontol. 2004;75:292-296.
   DOI: 10.1902/jop.2004.75.2.292
- Marcoli M, Candiani S, Tonachini L, Monticone M, Mastrogiacomo M, Ottonello A, Cervetto C, Paluzzi P, Maura G, Pestarino M, Cancedda R, Castagnola P. In vitro modulation of gamma amino butyric acid (GABA) receptor expression by stromal cells. Pharmacol Res. 2008;57(5): 374-82.

DOI: 10.1016/j.phrs.2008.03.012

- 23. Scotti R, Kantorski KZ, Monaco C, Valandro LF, Ciocca L, Bottino MA. SEM evaluation of in situ early bacterial colonization on a Y-TZP ceramic: A pilot study. Int J Prosthodont. 2007;20:419-422.
- Jiménez-Melendo M, Llena-Blasco O, Bruguera A, Llena-Blasco J, Yáñez-Vico RM, García-Calderón M, Vaquero-Aguilar C, Velázquez-Cayón R, Gutiérrez-Pérez JL, Torres-Lagares D. Mechanical behavior of single-layer ceramized zirconia abutments for dental implant prosthetic rehabilitation. J Clin Exp Dent. 2014;6(5): e485–e490.
  - DOI: 10.4317/jced.51442
- 25. Hisbergues M, Vendeville S, Vendeville P. Zirconia: Established facts and perspectives for a biomaterial in dental implantology J Biomed Mater Res B Appl Biomater. 2009;88(2):519-29. DOI: 10.1002/jbm.b.31147
- Tetè S, Mastrangelo F, Bianchi A, Zizzari V, Scarano A. Collagen fiber orientation around machined titanium and zirconia dental implant necks: An animal study. Int J Oral Maxillofac Implants. 2009;24(1):52-8.
- 27. Tatullo M, Simone GM, Tarullo F, Irlandese G, Vito D, Marrelli M, Santacroce L, Cocco T, Ballini A, Scacco S. Antioxidant and antitumor activity of a bioactive polyphenolic fraction isolated from the

- Brewing Process. Sci Rep. 2016;6: 36042. DOI: 10.1038/srep36042
- 28. Tatullo M, Marrelli M, Scacco S, Lorusso M, Doria S, Sabatini R, Auteri P, Cagiano R, Inchingolo F. Relationship between oxidative stress and "burning mouth syndrome" in female patients: A scientific hypothesis. Eur Rev Med Pharmacol Sci. 2012;16(9):1218-21.
- 29. Besinis A, Hadi SD, Le HR, Tredwin C, Handy RD. Antibacterial activity and biofilm inhibition by surface modified titanium alloymedical implants following application of silver, titanium dioxide and hydroxyapatite nanocoatings. Nanotoxicology. 2017;11(3):327-338.
- Scarano A, Piattelli A, Polimeni A, Di Iorio D, Carinci F. Bacterial adhesion on commercially pure titanium and anatase-coated titanium healing screws: An in vivo human study. J Periodontol. 2010;81(10): 1466-71.
- 31. Bernardi S, Bianchi S, Botticelli G, Rastelli E, Tomei AR, Palmerini MG, Continenza MA, Macchiarelli G. Scanning electron microscopy and microbiological approachfor the evaluation of microorganisms behavior on anatase titanium surfaces: vitro In studv. Morphologie. Dec 26; 2017. pii: \$1286-0115(17)30291-6.

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