Mechanical Property Assessment of Bone Healing around a Titanium–Zirconium Alloy Dental Implant

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ABSTRACT

Background: It has been reported that titanium–zirconium alloy with 13–17% zirconium (TiZr1317) implants show higher biomechanical stability and bone area percentage relative to commercially pure titanium (cpTi) grade 4 fixtures.

Purpose: This study aimed to determine whether the higher stability for TiZr1317 implants is associated with higher mechanical properties of remodeling bone in the areas around the implants.

Materials and Methods: This study utilized 36 implants ($n = 18$: TiZr1317, $n = 18$: cpTi), which were placed in the healed ridges of the mandibular premolar and first molar of 12 mini pigs ($n = 3$ implants/animal). After 4 weeks in vivo, the samples were retrieved, and resin-embedded histologic sections of approximately 100 μm in thickness were prepared. In order to determine the nanomechanical properties, nanoindentation ($n = 30$ tests/specimen) was performed on the bone tissue of the sections under wet conditions with maximum load of 300 μN (loading rate: 60 μN/s).

Results: The mean (± standard deviation) elastic modulus (E) and hardness (H) for the TiZr1317 group were 2.73 ± 0.50 GPa and 0.116 ± 0.017 GPa, respectively. For the cpTi group, values were 2.68 ± 0.51 GPa and 0.110 ± 0.017 GPa for E and H, respectively. Although slightly higher mechanical properties values were observed for the TiZr1317 implants relative to the cpTi for both elastic modulus and hardness, these differences were not significant ($E = p > 0.75$; $H = p > 0.59$).

Conclusions: The titanium–zirconium alloy used in this study presented similar degrees of nanomechanical properties to that of the cpTi implants.

KEY WORDS: animal model, biomechanics, bone, dental/endosteal implants, nanoindentation

INTRODUCTION

Commercially pure titanium (cpTi) has been widely used for oral implants due to its suitable biocompatibility and resistance to corrosion.[1–5] These properties contribute to the integration of the implants with the tissue as well as to an increased load bearing capability for subsequent prosthetic rehabilitation.[3,6] Although other commercially pure biomaterials have been proposed and

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tested (i.e., niobium and tantalum), it seems the commercially pure titanium presents the highest degree of biocompatibility.[7] Initially, dental implants were composed of grade I titanium bulk material.[8] However, due to its low yield strength, nowadays most of the commercially available oral implants are comprised of grade IV titanium, which has a yield strength approximately 180% higher.[9] It can be said that today grade IV cpTi is a well-documented successful implant material, which in general would function without complications.

However, it is a fact that not all implants function as anticipated and at times generate complications such as fixture fracture and/or marginal bone loss. Several factors, such as patient parafunctional habits, misfit of the prosthesis, fatigue of the material, and design of the implant, are said to be associated with unfortunate clinical outcomes.[9–11] For instance, in the mandibular anterior region, the oral anatomy might require a reduced implant diameter.[12] In such cases, clinicians are suggested to select a biomaterial that can withstand the high level of forces, which may be generated by the factors mentioned above. For this purpose, biphasic ($\alpha + \beta$) titanium alloys, such as Ti6Al4V, present higher mechanical properties under cyclic loading relative to single α phase cpTi alloys.[5] Thus, nowadays, some implant manufacturers offer titanium alloys as implant biomaterials.[13] However, reports speculating about the potentially negative biocompatible properties of its components along with the difficulties to obtain an optimal surface roughness have been an obstacle for Ti6Al4V to become a prevailing selection.[14,15] For these reasons, the development of alternative titanium alloys free from aluminium and vanadium elements but with similar level of high mechanical properties is of great interest.[5] For this purpose, titanium alloy with 13–17% zirconium TiZr1317 has been introduced and reported to be a promising alternative to Ti6Al4V.[16] This biomaterial has mechanical properties comparable with those of Ti6Al4V and presents a similar biologic response to that of cpTi fixtures.[17,18] Furthermore, due to its monophasic α structure, TiZr1317 has been said to develop microtopographies by acid etching and sand blasting similar to that of cpTi implants.[18]

It has been reported by Gottlow and colleagues that TiZr1317 implants placed in mini pigs presented significantly higher removal torque values compared with those of cpTi implants with same macrogeometry. Histomorphometric measurements showed a statistically significant higher bone area within the chamber for the TiZr1317 implants than the titanium (Ti) implants.[17] Interestingly, the histomorphometric bone-to-implant contact percentage presented no significant differences. It could be expected that further investigations demonstrate that the mechanical properties of the newly formed bone would show similar levels of tissue mineralization between TiZr1317 and cpTi specimens.

Thus, in order to determine the biomechanical properties within the bone formed around two implants with different bulk materials with similar macrogeometric design and surface topography, the elastic modulus and hardness of newly formed bone in proximity with the implant surface were investigated with a nanoindentation technique.

MATERIALS AND METHODS

In Vivo Protocol

The in vivo animal trial protocol was reported in details in the previous work by Gottlow and colleagues.[17] In summary, 12 adult female Göttingen mini pigs (Ellegaard, Denmark), 14 to 16 months old and around 20 to 29 kg in weight were used. For each pig, the mandibular premolars (P1, P2, and P3) and the first molar (M1) were extracted prior to the study. After 3 months of healing, three bone chamber implants were implanted in one hemi-mandible of each mini pigs ($n = 36$ in total). Eighteen fixtures were TiZr1317 (Roxolid, Institut Straumann AG, Basel, Switzerland) and the other 18 were cpTi grade 4 implants (Institut Straumann AG). The two types of implant benefited of a sandblasted acid-etched hydrophilic surface (SLActive, Institut Straumann AG). At the end of the 4-week healing period, the animals were sacrificed after general anesthesia with an intracardiac injection of a lethal dose of sodium pentobarbital. Immediately after sacrifice, the mandibles were excised, and the hemi-mandibles were sectioned using a bandsaw and immersed in 4% formaldehyde solution for histologic processing.

Sample Preparation

For the current study, the remaining half of the bone-implant tissue resin blocks, which the first half was used for the histological processing by Gottlow and colleagues,[17] were further cut into approximately 200 μm sections along the bucco-lingual long axis using a precision diamond saw (Isomet 2000, Buelher, Lake Bluff, IL, USA) and glued to acrylic plates with acrylate-based cement. After a setting time of 24 hours, specimens were ground (400 to 2400 grit silicon carbide abrasive paper) and polished down to a final thickness of 150 µm (diamond suspensions of 9 to 1 µm particle size) (Buehler). Prior to mechanical testing, all specimens were stored in water for 24 hours.

Nanoindentation Testing

Nanoindentation ($n = 30$/specimen) was performed using a nanoindenter (Hysitron TI 950, Minneapolis, MN, USA) equipped with a Berkovich diamond three-sided pyramid probe. A wax chamber was created above the acrylic plate around the implant-in-bone perimeter, so that tests were performed in water.[19,20] For each specimen, mechanical testing was conducted in the healing chamber regions (Figure 1). Bone tissue was detected by imaging under the light microscope (Hysitron TI 950), and indentations were randomly performed in selected areas where the bone existed and made sure that the values obtained were not from other tissue or resin itself (Figure 1). A loading profile was developed with a peak load of 300 µN at a rate of 60 µN/s, followed by a holding time of 10 s and an unloading time of 2 s. The extended holding period allows bone to relax to a more linear response, so that no tissue creep effect occurred in the unloading portion of the profile (ISO 14577-4). Therefore, from each indentation, a load-displacement curve was obtained that have been described in previous publications.[19,20] From each curve, reduced modulus (GPa) and hardness (GPa) of bone tissue were computed using Hysitron TriboScan software and its elastic modulus E (GPa) was calculated as follows:

$$\frac{1}{E_r} = \frac{1-\nu^2}{E} + \frac{1-\nu_i^2}{E_i}$$

where Er is the reduced modulus (GPa), ν (0.3) is the Poisson's ratio for cortical bone, Ei (1140 GPa) and νi (0.07) are the elastic modulus and Poisson's ratio for the indenter.[21–23]

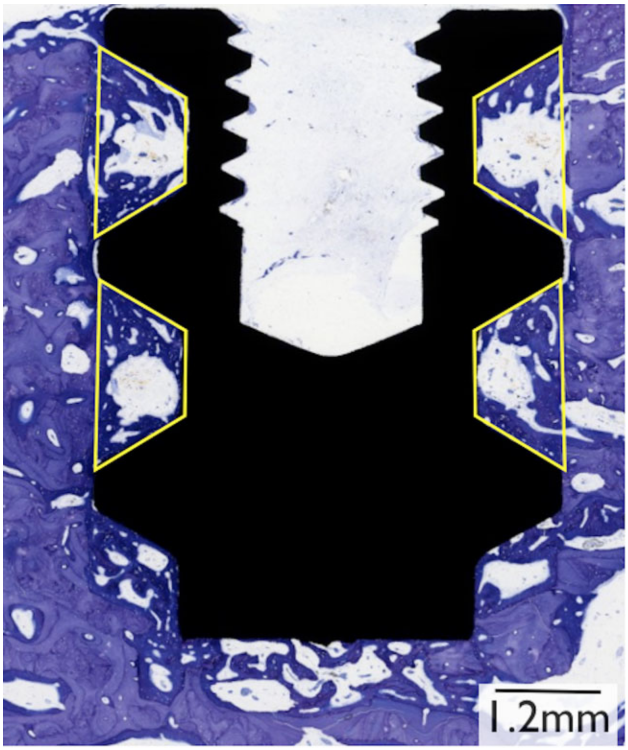

Figure 1 Representative stained section of an implant in bone sample depicting the healing chamber implant design (chambers delineated by the yellow boxes).

Statistical Analysis

Statistical software (SPSS, IBM, Armonk, NY, USA) was employed so that linear mixed models were performed to determine the influence of different implant (TiZr1317 vs. Ti groups) and position (anterior, mid, posterior) on ranked elastic modulus and ranked hardness values. Statistical significance was set at 95% level of confidence.

RESULTS

The overall elastic modulus and hardness values ranged from 0.46 to 9.41 GPa and from 0.005 to 0.014 GPa (overall mean recorded was 0.111 GPa), respectively. The mean (± standard deviation) elastic modulus and hardness for the cpTi group were 2.68 ± 0.51 GPa and 0.110 ± 0.017 GPa, respectively. For the TiZr1317 group, the mean (± standard deviation) elastic modulus and hardness were 2.73 ± 0.50 GPa and 0.116 ± 0.017 GPa, respectively. While slightly higher mechanical properties values were observed for the TiZr1317 implants relative to the cpTi for both elastic modulus and hardness, no significant differences were found between the two groups (elastic modulus [E]: $p > 0.75$; hardness [H]: $p > 0.59$). Furthermore, no effect of implant position in the mandible (i.e., anterior, mid, and posterior) was observed (E: $p > 0.41$; H = $p > 0.95$).

Summary of the statistical results for both the elastic modulus values and ranked elastic modulus and for both hardness values and ranked hardness obtained for

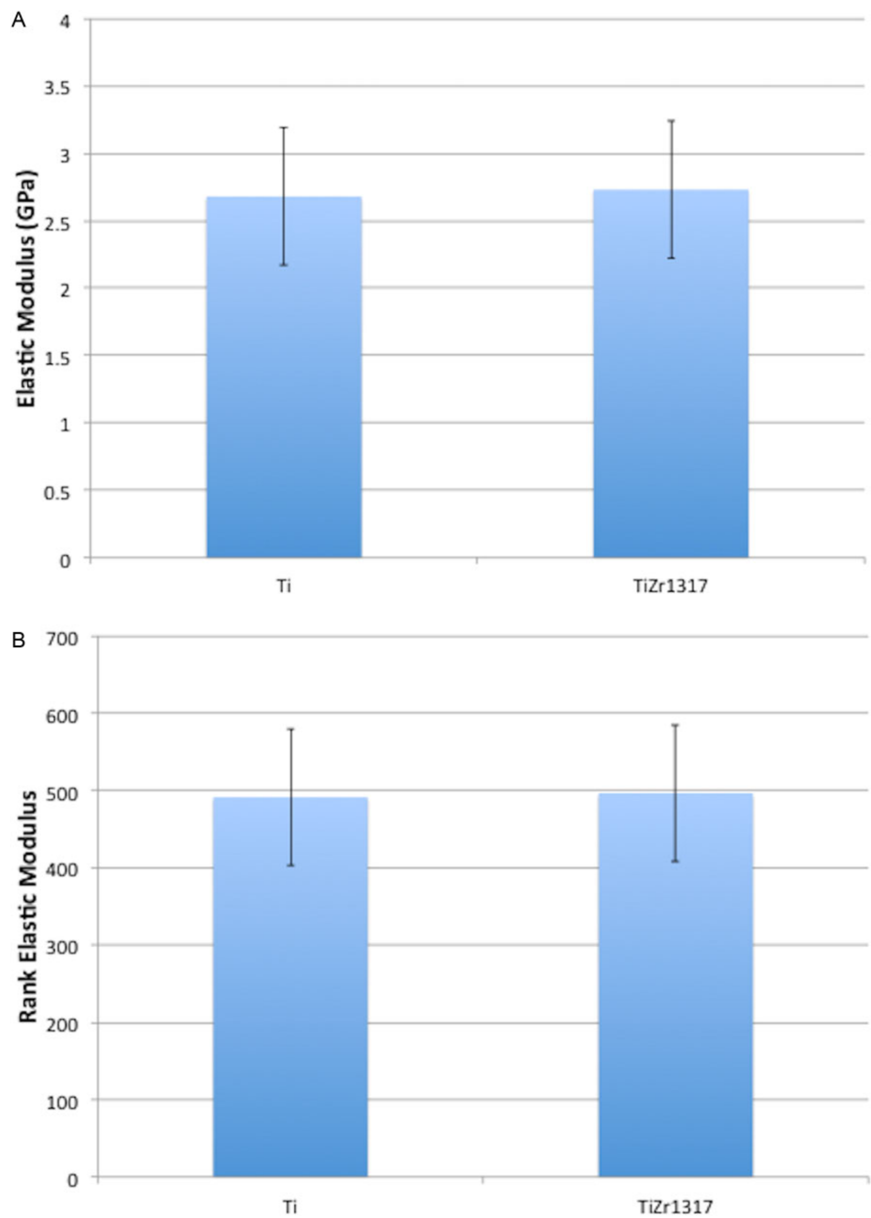

Figure 2 Summary statistics (mean ± standard error) for both groups' (A) elastic modulus and (B) rank elastic modulus.

the two different groups (i.e., cpTi and TiZr1317) are shown in Figures 2 and 3, respectively.

DISCUSSION

This study focused on the comparison of the mechanical properties of bone around commercially pure titanium grade 4 and titanium-zirconium (TiZr1317) alloy implants using the nanoindentation technique. The results indicated that the two different implant biomaterials did not affect either the elastic modulus or hardness of the surrounding bone, suggesting that the level of bone mineralization inside the experimental chambers occurring at 4 weeks in vivo in the miniature pig were similar. Since the bone of the mini pig has a turn over similar, but slightly faster than the human bone,[24] 4 weeks of healing represents a rather early time point during osseointegration. This time point was selected since in the previous study by Gottlow and colleagues (2012), the removal torque results presented significantly higher values for the TiZr1317 alloy implants compared with the titanium implants, but the histomorphometric values were comparable.[17] We hypothesized that there may be differences in the mineralization levels that is difficult to capture with the histomorphometry

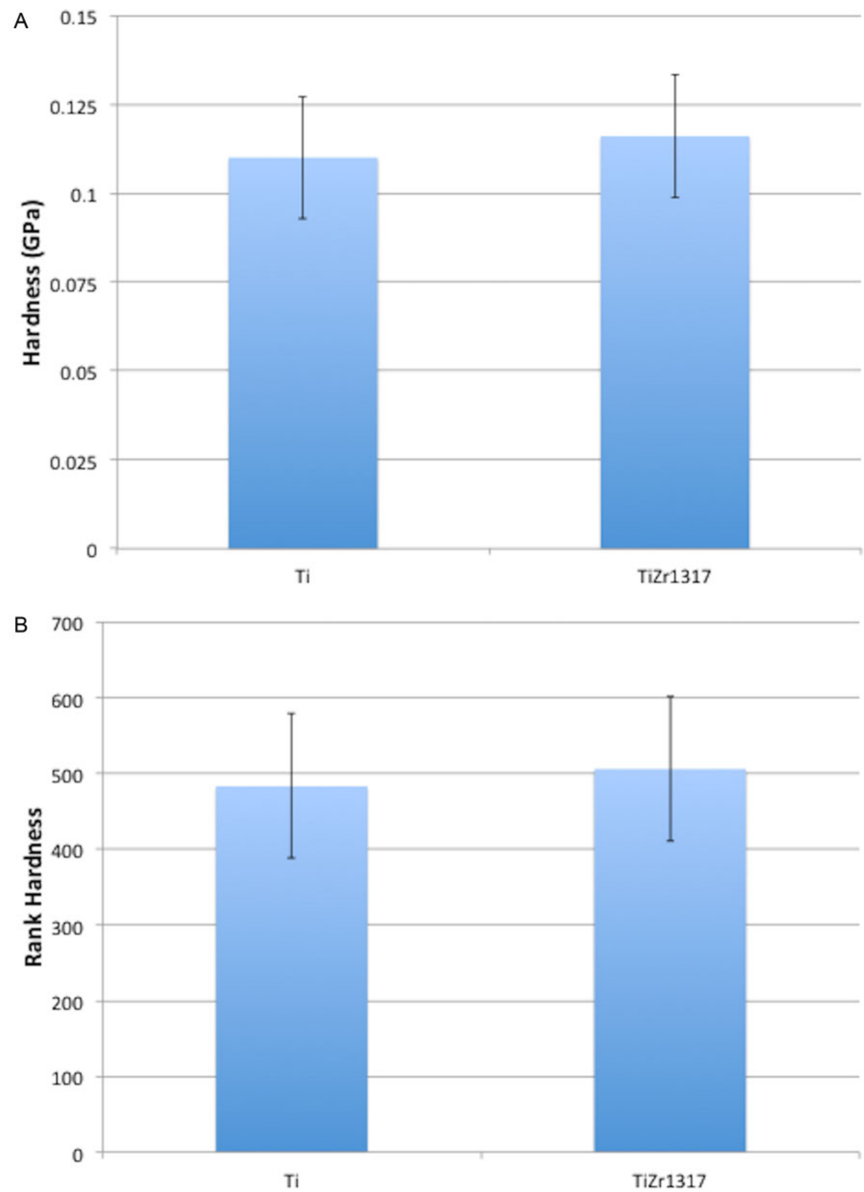

Figure 3 Summary statistics (mean ± standard error) for both groups' (A) hardness and (B) rank hardness.

and thus conducted nanoindentation using the sections from the same study.

TiZr1317 presents significantly better mechanical properties relative to pure titanium.[25] Thus, it was critical to observe how different materials' mechanical properties would affect the mechanical performance of bone. To our knowledge, this is the first study focusing on the comparison of the mechanical properties of bone between two different titanium-based implants. The results presented no significant differences in either the elastic modulus and hardness, although in the previously published study by Gottlow and colleagues, bone area and removal torque resistance were significantly higher for the TiZr1317 than for pure titanium implants.[17] This implies that better biomechanical resistance may be due to the higher bone area, which embedded the implant within newly formed bone. Although no significant differences were found between the two implant materials in bone-to-implant contact in that study, the increased bone area around the implant may provide firmer support. Therefore, the results of the current study suggest that the higher implant stability may be due to higher bone area rather than faster mineralization of bone. The higher bone area could be due to the different mechanical properties of the two implant materials and/or to the surface topography, which is similar but not identical between the two types of implants at both micro- and nanolevels. This has been confirmed in a recent study by Wennerberg and colleagues,[26] where they indicated that the two implant surfaces presented different topographical values and that surfaces do not present identical topography since the material properties are different.

Nanoindentation of bone around implants has the potential to elucidate the qualitative aspects of osseointegration.[20] The nanomechanical properties of the tissue surrounding implants were first investigated by Butz and colleagues, who focused on the effect of different surface topographies on titanium implants.[6] The study, performed on a rat model, revealed that textured surfaces were surrounded by harder bone relative to smoother surfaces at different time points. This suggests that the so-called moderately rough surfaces not only have a mechanical interlocking effect,[27] but also act to stimulate cellular response, enhancing direct cell attachment to the implant and mineralization of newly formed bone.[28]

The nanoindentation technique is a suitable method to take into account the anisotropic organization of bone[23] and map the range of mechanical properties of bone around implantable devices. With this technique, the process of bone mineralization relative to implant design variations (e.g., instrumentation, bulk, and surface) can be evaluated. Since the current study conducted the nanoindentation for both control and test groups under wet conditions, the results cannot be directly compared with the ones conducted under dry conditions, which has indeed shown consistent outcomes.[29] However, since the test and control specimens were treated in the same manner, both groups were compared on a relative basis as performed in our previous investigations.[19,20]

CONCLUSION

The results of the current study indicated that no significant differences in bone mechanical properties were detected between cpTi grade 4 and TiZr1317 at 4 weeks healing in the mini pig mandible model.

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