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A new cervical implant design compared with standard design in order to increase peri-implant hard and soft tissue behavior: histomorphometric and histological study in dogs

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Date:

Accepted 1 July 2016

To cite this article:

Calvo-Guirado JL, Maté-Sánchez de Val JE, Delgado-Ruiz RA, Fernández Domínguez M, Orlato Rossetti PH, Gehrke SA. A new cervical implant design compared with standard design in order to increase peri-implant hard and soft tissue behavior: histomorphometric and histological study in dogs. *Clin. Oral Impl. Res.* 00, 2016, 1–8
 doi: 10.1111/clr.12978

Key words: crestal bone, hard and soft tissue, new implant design

Abstract

Objective: The aim of this study was to evaluate a new design of the cervical portion of dental implant with the objective to increase the volume of peri-implant tissues in the crestal area.

Materials and Methods: Forty-eight tapered dental titanium implants with internal conical connection were implanted in healed alveolar sites of six dogs. Twenty-four conventional implants design (C1 implant) formed the control group, and 24 new implant design (V3 implant) formed the test group. The groups were randomized. Histological, histomorphometric, and implant stability quotient were performed. After 12 weeks of healing period, histomorphometric analyses of the specimens were carried out to measure the crestal bone level values and the tissue thickness in the cervical implant portion. The data were compared using statistical tests ($\alpha = 5\%$).

Results: The mean of the measurements in the buccal and lingual aspects measured of crestal bone level was 0.31 ± 0.24 mm and 0.30 ± 0.19 mm in the control group, respectively, and 0.71 ± 0.28 and 0.42 ± 0.30 mm in the test group, respectively, whereas the mean of the tissue thickness was 1.63 ± 0.33 mm and 2.04 ± 0.23 mm in the control group, respectively, and 2.11 ± 0.35 mm and 2.51 ± 0.41 mm in the test group.

Conclusions: Within the limitations of this study, our findings suggest that more thickness of peri-implant hard and soft tissues may be expected in this new implant design. However, the control group with traditional implant design was found to have more height values of the crestal bone compared with new V3 implants.

Actually, the replacement of tooth loss by implant-supported prosthesis has been widely used in the dentistry practice, mainly due the predictability showed in the last decades. However, some factors may complicate the indication and execution of this treatment modality, such as the quantity of bone tissue, because of a progressive involution of the alveolar bone in both the horizontal and the vertical dimensions after the tooth loss (Botticelli et al. 2004; Araújo & Lindhe 2005) and because the most rapid reduction in the alveolar bone after tooth extraction occurs during the first months (Araújo et al. 2005; Schropp et al. 2009). In the recent past, the control of these installed and dental implants was considered successful when properly corresponded in function, but, in accordance with the literature, the mean bone loss over the first year post-implant is 0.9–1.5 mm, and

the subsequent loss per year is around 0.1 mm (Albrektsson et al. 1994). However, this concept has been reconsidered during the last years due to the growth of the aesthetic requirements. So, various techniques and materials have been developed to limit resorption of the alveolar ridge after tooth extraction (Brandam et al. 2015) and after implant placement (Hermann et al. 2007), and results from using them have been promising.

With regard to the techniques, some authors suggested that immediate implant placement may counteract the bone remodeling process and preserve the dimension of the alveolar ridge (Paolantonio et al. 2001). However, multiple animal investigations have failed to support this hypothesis (Araújo et al. 2005; Botticelli et al. 2006). In this sense, Araújo et al. (2005, 2006) found a

pronounced resorption of the buccal and lingual bony walls after immediate placement in fresh extraction sockets. In long-term observations, no significant differences in the success and aesthetic outcomes have been reported between immediate and delayed implants (Grunder et al. 1999; Mangano et al. 2012; Chen & Buser 2014). Relative to the materials, new micro- and macro-designs were developed with switching platform (Calvo-Guirado et al. 2014; Romanos & Javed 2014), cervical micro-threads (Calvo-Guirado et al. 2015, 2016), smooth or rough surface (Hermann et al. 2011; Schwarz et al. 2014), implants in one or two pieces (Hermann et al. 2001), internal or external connection of abutment (Schwarz et al. 2014).

In aesthetics sectors that are rehabilitated with dental implants, any loss of peri-implant tissue dimension (vertical or horizontal) can influence the results and can be considerable (Palacci 2004). In this way, the objective of the present study was to evaluate a new cervical implant design with regard to the healing of peri-implant tissues during osseointegration of dental implants placed in healed sites using a mandible dog model.

Material and methods

Implants characteristics and groups

This experimental animal study utilized 48 tapered titanium implants, with the same surface characteristics (sandblasted and acid etching) but with different macro-design. The 48 conical implants were divided into two groups ($n = 24$ per group): control group, C1 implant (MIS, Bar lev Industrial Park, Israel) with conventional cervical implant design (circular); and a test group, in which a new cervical implant design (triangular) was tested, called V3 implant (MIS). The characteristics of each implant are presented in Fig. 1.

Surgical procedure and animals care

Six American foxhound dogs of approximately 1 year of age were used in this study. The Ethics Committee for Animal Research at The University of Murcia (Spain) approved the study protocol, which followed the guidelines established by the European Union Council Directive of February 2013 (R.D.53/2013). Clinical examination determined that all animals were in good general health; moreover, all animals presented intact maxillae, without occlusal trauma or mucosal lesions.

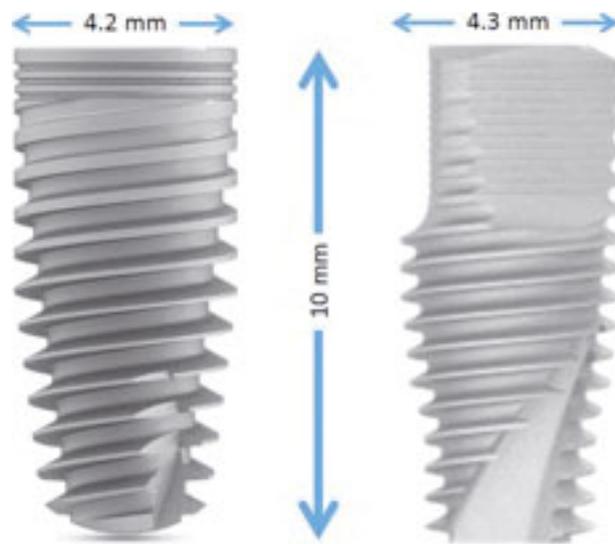


Fig. 1. Images of the implants used in this study.

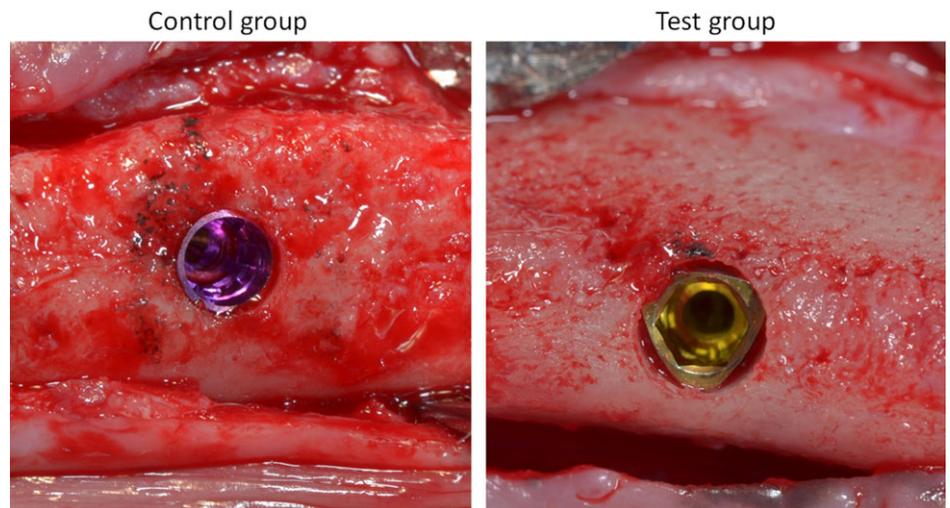


Fig. 2. Images of the circular and triangular head implants installed in healed sites.

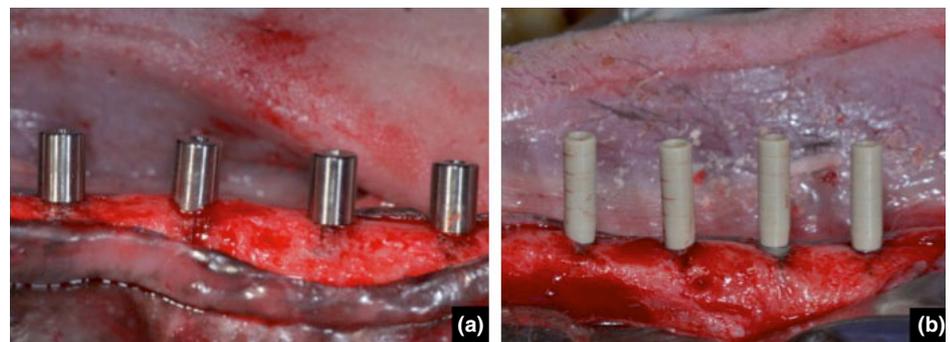


Fig. 3. (a) Image of the circular and triangular head implants with titanium abutments, (b) peek abutments screwed on top of both implants.

The animals were pre-anesthetized with acepromazine 0.12%–0.25 mg/kg, buprenorphine 0.01 mg/kg, and medetomidine 35 mg/kg. This mixture was injected intramuscularly in

the femoral quadriceps. Animals were then taken to the operating theater, where an intravenous catheter was inserted into the cephalic vein, and propofol (0.4 mg/kg/min)

was infused. Conventional dental infiltration anesthesia (articaine 40 mg, 1% epinephrine) was injected at the surgical intraoral sites. All procedures were carried out under the supervision of a veterinary surgeon. Initially, an impression of each hemimandible was performed to make a surgical guide to indicate the implant position, which was predetermined to correspond with the distal root and the center of the crown teeth. Sixty days before the surgery, the bilateral mandibular premolars (P2, P3, P4) and molar (M1) were extracted to heal the alveolus sites. For this, the teeth were sectioned in a buccolingual direction using a tungsten carbide bur so that the roots could be extracted individually without damaging the remaining bony walls. In the second surgery to place the implants, full-thickness mucoperiosteal flaps were inserted and eight implants were installed in each animal. The distribution was randomized in each hemimandible (www.randomization.com). All implants of both groups were positioned in the crestal bone level (Fig. 2), and two different abutments provisional peek and titanium abutments (MIS) were screwed on each implant and cut at 1-mm infraocclusion (Fig. 3a,b). The reason of using two different abutments is to control soft tissue healing in order to measure gingival length, and to heal and protect crestal bone resorption.

The flaps were closed using single sutures (Silk 4.0, Lorca Marin, Murcia, Spain). After the surgical procedures, animals received antibiotic treatment (amoxicillin 500 mg, twice a day) and analgesics (Ibuprofen 600 mg, three times a day) via the systemic route. Moreover, dogs were fed a soft diet for 7 days, and plaque control was maintained by the application of Sea4 (Sea4 teeth, Blue Sea Laboratories, Alicante, Spain) spray. Wounds were inspected daily for clinical postsurgical complications. Two weeks after surgery, sutures were removed and digital Rx was taken.

All animals were sacrificed at 12 weeks after the implant insertion by means of an overdose of Pentothal Natrium® (Abbot Laboratories, Madrid, Spain) and perfused through the carotid arteries with a fixative containing 5% glutaraldehyde and 5% formaldehyde. After sacrifice, digital Rx was taken as the last radiographic evaluation follow-up.

Resonance frequency analysis

All animals were used for resonance frequency analysis (RFA) to measure the implant stability. A Smartpeg™ (Integration Diagnostics AB, Göteborg, Sweden) was screwed into each implant and tightened to approximately

5 N. The transducer probe was aimed at the small magnet at the top of the Smartpeg at a distance of 2 or 3 mm and held stable during the pulsing until the instrument beeped and displayed the ISQ value. For RFA, the implants were measured immediately after the installation and 12 weeks after the implant insertion during removal. The implant stability quotient (ISQ) values were measured by Osstell™ Mentor (Integration Diagnostics AB). The ISQ values were measured in two directions mesial to distal and buccal to lingual, and an average of each sample was determined. A mean value was calculated from the measurements taken parallel to the long axis of the mandible.

Histological preparation and histomorphometric analysis

Specimens were washed in saline solution and fixed in 10% buffered formalin and sent for processing at the Laboratory of Ucam-Biotecnos (Murcia, Spain). Specimens were dehydrated in ascending series of alcohol rinses and embedded in a glycol methacrylate resin (Technovit 7200 VLC; Kulzer, Wehrheim, Germany). After polymerization, the

specimens were sectioned along its longitudinal axis with a high-precision diamond disk in the IsoMet® 1000 (Buehler, Lake Bluff, IL, USA), at about 150 µm down to 30 µm. A total of two slides were obtained for each implant. The slides were stained with Picrosirius red stain (Polysciences Inc., Warrington, FL, USA) and observed in a normal transmitted light microscope and a polarized light microscope (Nikon, Tokyo, Japan). Buccal bone wall level in comparison with lingual bone wall resorption was expressed as a linear measurement from the implant shoulder (A) to the first implant-to-bone contact (B). The thickness of buccal and lingual tissue was measured in a line corresponding with the implant shoulder level (A), from the implant surface (C) to the more external portion of the epithelium (D). Figure 4 shows the scheme used to the measurements. All measurements were taken by an expert professional in histology (SG).

Metric evaluation of the predetermined parameters was carried out using a light microscope (Nikon, Tokyo, Japan) connected to a high-resolution video camera (3CCD, JVC KY-F55B, JVC®, JVC, Yokohama, Japan). After digitizing the phase of each specimen under light the microscope, all proposed details were measured in the images using the program Image Tool version 5.02 for Microsoft Windows™ (UT Health Science Center School of Dentistry, San Antonio, TX, USA).

Statistical analysis

In order to evaluate the data, implant was used as the statistical unit for all measurements. Statistical Package at the University of Murcia (SPSS 20.0, SPSS Inc., Chicago, IL, USA) program was used for the statistical analyses. Bruner–Langer test was used to compare crestal bone loss in all groups.

Furthermore, a descriptive statistical methods (mean, SD, median), in comparison with quantitative data, and *t*-test were used to compare the two groups when the parameters were of normal distribution among groups at the buccal or lingual sites for A–B and C–D parameters, respectively. Nonparametric Mann–Whitney *U*-test was used for

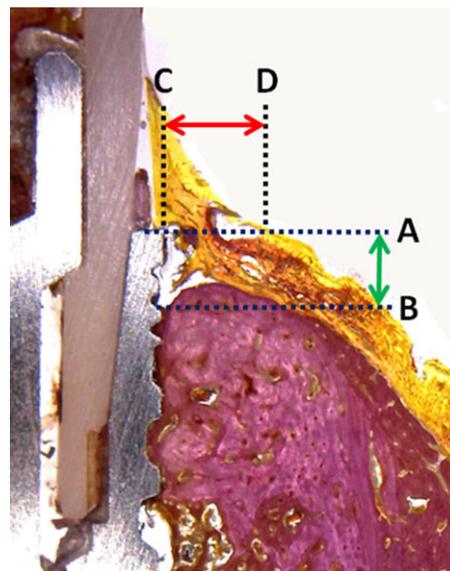


Fig. 4. Scheme to show the measurement in the crestal tissues (bone position A to B and tissue thickness C to D) in the buccal and lingual aspects of each implant.

Table 1. Bruner–Langer test of ISQ analysis and measurements at baseline (initial) and at 12 weeks. Results as mean and medians. Nonparametric Bruner–Langer test was used to compare intergroups. (*) Significant differences, $P < 0.05$

| ISQ value | Baseline | | 12 weeks | | P value |
|----------------------|--------------|--------|---------------|--------|---------|
| | Mean ± SD | Median | Mean ± SD | Median | |
| Control group | 70.05 ± 3.41 | 69.91 | 73.56 ± 4.23* | 73.39 | 0.09 |
| Test group | 71.18 ± 3.55 | 70.79 | 73.22 ± 4.34* | 73.20 | 0.14 |
| P value (intergroup) | 0.054 | | 0.061 | | |

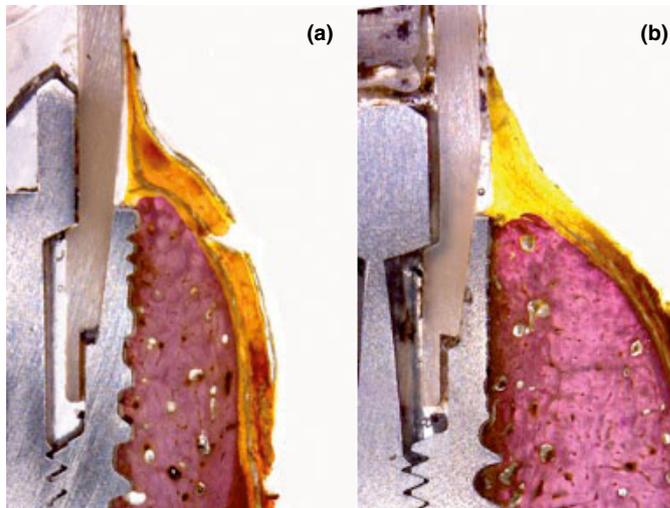


Fig. 5. Histological images of the implants or the control group (a) and test group (b) in the buccal position, which presented more significant difference between C1 and V3 implants.

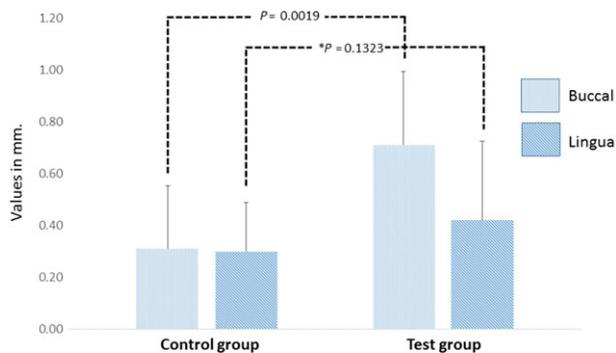


Fig. 6. Bar graph of the values and statistical analysis of the bone height measured from the implant shoulder to the first bone-to-implant contact.

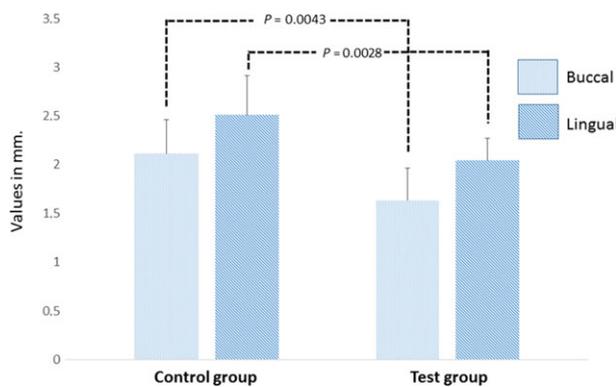


Fig. 7. Bar graph of the values and statistical analysis of the tissue thickness measured from the implant shoulder to the external portion (epithelium) of the mucosa.

Table 2. Crestal bone loss at 12 weeks. (a) Significant difference in comparison between control and test. (b) Significant difference in comparison between B and L. Nonparametric Bruner–Langer test was applied. Values recorded as mean ± SD and median (x)

| | AB distance | | | CD distance | | |
|---------------|---|---------------------------------------|-----------|---|---------------------------------------|-----------|
| | B | L | P value | B | L | P value |
| Control group | 0.30 ± 0.22 (0.313) | 0.30 ± 0.19 (0.301) | (b) 0.063 | 1.60 ± 0.32 (1.612) | 2.05 ± 0.20 (2.034) | (b) 0.051 |
| Test group | 0.75 ± 0.29 ^{(a),(b)} (0.753) | 0.45 ± 0.28 ^(a) (0.450) | (b) 0.025 | 2.10 ± 0.35 ^{(a),(b)} (2.078) | 2.50 ± 0.41 ^(a) (2.479) | (b) 0.017 |
| P value | (a) 0.032 | (a) 0.011 | | (a) 0.018 | (a) 0.041 | |

comparison, assuming a level of significance of 95% ($P < 0.05$).

Results

The surgical sites healed uneventfully. All animals presented appropriate healing during the first week following the surgical procedure. Post-surgical inspections for 2 weeks postoperatively indicated the absence of infection or inflammation. All implants presented osseointegration in both periods proposed and were available for histological analysis.

Regardless to the RFA, all data (mean, SD, median and P values) of resonance frequency values for the two investigated implant designs are summarized in Table 1. The Bruner–Langer test was used to compare the values between the groups in the 12-week period and no statistical differences within and between groups were observed, with significance set at $P < 0.05$.

Direct contact was observed between living bones and all implants without the presence of soft tissues were observed in both groups. However, during the healing period, the crestal areas were accompanied by decreases in the dimensions of the buccal as well as the lingual bone walls in different proportions for each group. For all implants, keratinized oral epithelium was continuous with junctional epithelium facing the implants and the healing screws. Subjacent connective tissue with a dense network of collagen fibers was observed. After evaluating all measurements, a mean of the buccal and lingual aspects measured from each group was taken and the distance from the top of the implant collar (line A) to the first bone-to-implant contact (line B) was 0.31 ± 0.24 mm in buccal and 0.30 ± 0.19 mm in lingual for the control group, and 0.71 ± 0.28 mm in buccal and 0.42 ± 0.30 mm in lingual for the test group. These data showed statistically significant differences at 12 weeks among the groups in the buccal aspect ($P = 0.0019$) and no differences were found in the lingual aspect ($P = 0.132$), whereas the mean of the tissue thickness from the top of the implant shoulder (line C) to the more external portion of the tissues (line D) was 1.63 ± 0.33 mm in buccal and 2.04 ± 0.23 mm in lingual for the control group, and 2.11 ± 0.35 mm in buccal and 2.51 ± 0.41 mm in lingual for the test group. These data showed statistically significant differences at 12 weeks among the groups in the buccal aspect ($P = 0.0043$) and the lingual aspect ($P = 0.0029$). Figure 5 shows the images of the both groups in the

Table 3. Radiographic evaluation of marginal bone loss in both types of implants at baseline, 15 days, and 12 weeks. Mean value (SD) at 12-month follow-up period

| Radiographic crestal bone loss | Baseline triangular V3 Mean \pm SD (mm) | Baseline circular C1 Mean \pm SD (mm) | 15 days triangular V3 Mean \pm SD (mm) | 15 days circular C1 Mean \pm SD (mm) | 12 weeks triangular V3 Mean \pm SD (mm) | 12 weeks circular C1 Mean \pm SD (mm) | P value |
|--------------------------------|---|---|--|--|---|---|---------|
| Mesial site | 2.54 \pm 0.2 | 2.36 \pm 0.8 | 2.67 \pm 1.5 | 2.43 \pm 1.3 | 2.79 \pm 0.7 | 2.51 \pm 0.8* | 0.038* |
| Distal site | 2.61 \pm 0.4 | 2.45 \pm 0.5 | 2.69 \pm 0.9 | 2.50 \pm 1.1 | 2.78 \pm 1.1 | 2.53 \pm 1.7* | 0.049* |

*Significant differences, $P < 0.05$.

buccal aspect. The illustrative distribution of collected data is shown as bar graphs in Fig. 6 for the crestal bone height and in Fig. 7 for the tissue thickness measurements.

Titanium and peek healing abutments did not have any statistical significance influence related to crestal bone resorption and gingival healing (Table 2).

Radiographic evaluation

Crestal bone resorption on mesial and distal sites in C1 and V3 implants was evaluated. More crestal bone resorption was found from the baseline in mesial (2.79 ± 0.7 mm) and distal sites (2.78 ± 1.1 mm) in triangular V3 implants compared with circular C1 implants in mesial (2.51 ± 0.8 mm) and distal sites (2.53 ± 1.7 mm), and these data are summarized in Table 3.

Figures 8 and 9 explain the crestal bone resorption in triangular and circular implants with titanium and peek abutments. More crestal bone resorption was observed in V3 implants compared with C1 implants at 12 weeks follow-up with titanium abutments. Furthermore, the peek abutments maintained crestal bone resorption in both different implant designs.

Bruner-Langer test identified no statistical significant differences among groups for each site and parameter regarding crestal bone loss and tissue thickness ($P < 0.001$). Crestal bone loss was higher for the test group (V3 implants) at the buccal site compared with control sites (C1 implants). On the other hand, tissue thickness was higher for the test group at both buccal and lingual sites. The correlation coefficients are shown in Table 3. A strong and statistically significant correlation was only found between A-B and C-D parameters for the control group at the lingual site, as shown in Fig. 10.

Discussion

The conservancy of bone around the implant especially in the buccal plate plays a crucial role on esthetics. Resorption of buccal plate may lead to exposed threads, thus affecting the esthetic of the treatment, even if prostheses are not still connected (Boquete-Castro

et al. 2015). In an effort to stabilize and/or control bone crest height relative to the apical portion of the implant, several new techniques and macro- and micro-designs of implants have been proposed (Bratu et al. 2009; Nickenig et al. 2009; Hermann et al. 2011). Additionally, a wide range of modifications in the implant design have been developed by the companies to enhance the effect of the threads and improving the initial implant stability. However, very few have been scientifically documented. Thread patterns in dental implants currently range from micro-threads near the neck of the implant to broad macro-threads on the mid-body and a variety of altered pitch threads to induce self-tapping and bone compression (Binon 2000; Abuhussein et al. 2010). Then, the present Investigation showed the tissue behavior

after 12-week healing period which affected both buccal and lingual crestal bone and the tissue thickness in the portion corresponding to a new cervical implant design, and it was also found to be a possible alteration in the stability of implants.

Using resonance frequency analysis (RFA), it is now possible to measure the ISQ at any time during the course of implant treatment and loading (Meredith et al. 1996; da Silva Neto et al. 2013; Gehrke et al. 2016). Using ISQ values as a parameter to assess the tested implants is noninvasive. It was reported to be a reliable and accurate method for the early assessment of implant stability that is related to the bone-implant interface (Meredith 1998; Huang et al. 2003). Thus, in this study there was a concern in assessing the behavior of the new implant design, with regard to the

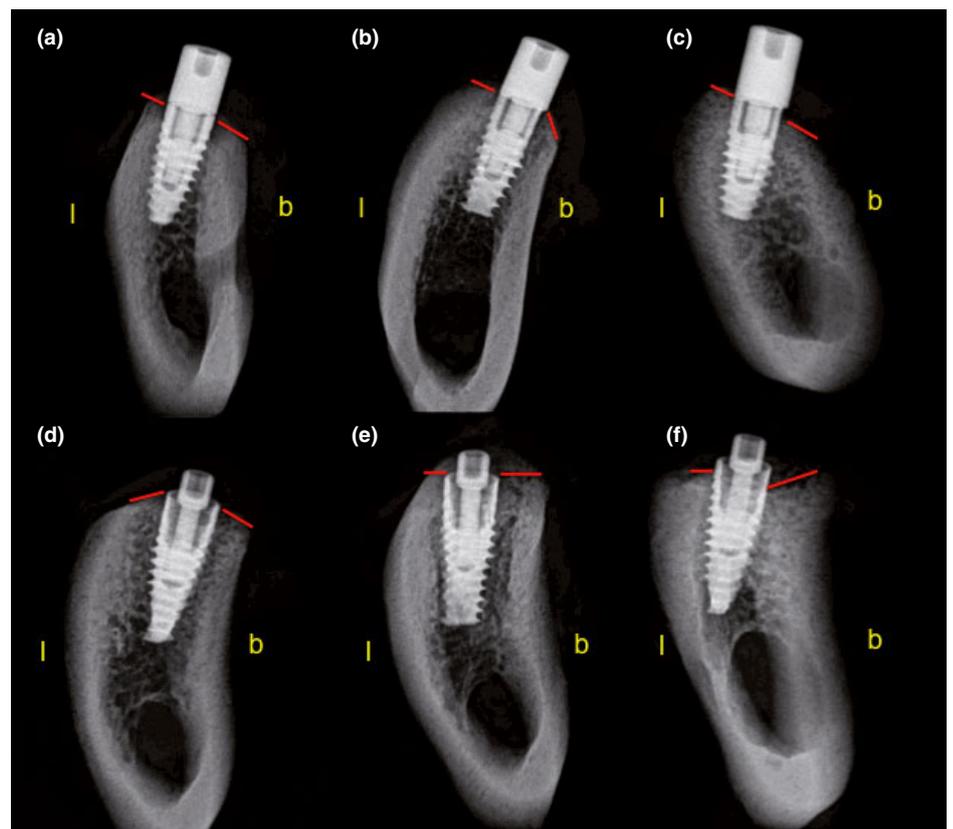


Fig. 8. Crestal bone resorption in triangular implants with titanium abutments, (a) triangular v3 implant at baseline, (b) triangular v3 implants with less buccal bone resorption at 15 days, (c) more resorption at 12 weeks in triangular v3 implants, (d) triangular v3 implants with peek abutments at baseline, (e) v3 implant with peek abutment at 15 days with less bone resorption and (f) triangular v3 implant with peek abutment showed buccal and lingual bone resorption at 12 weeks.

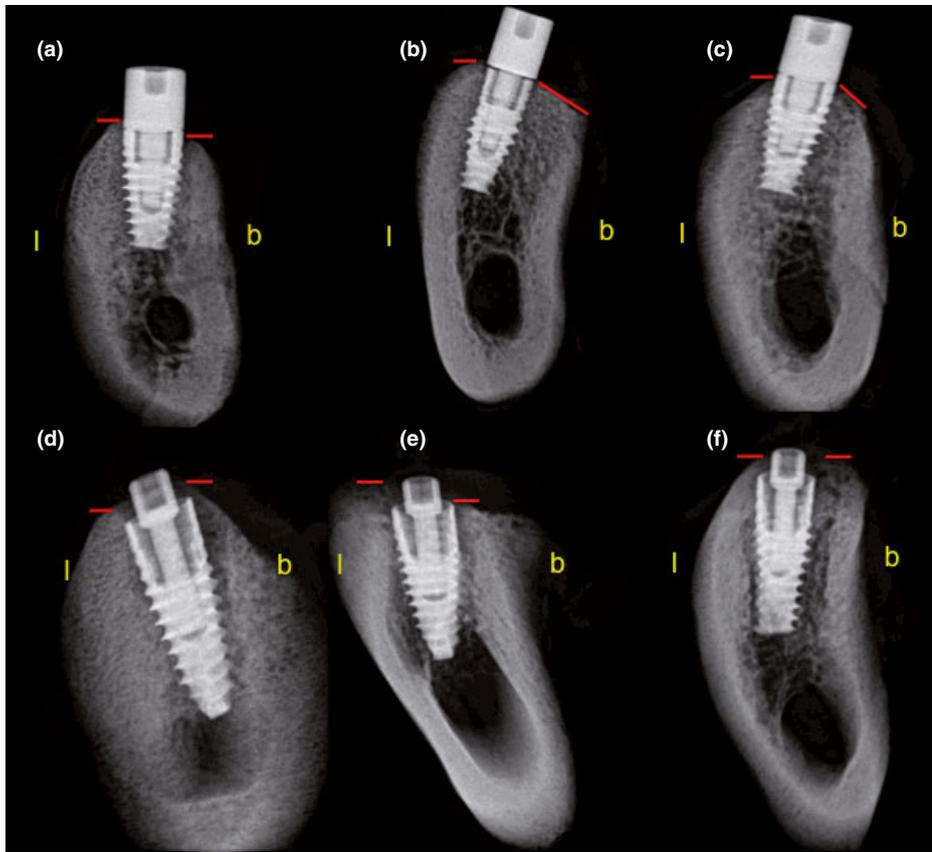


Fig. 9. Crestal bone resorption in circular standard C1 implants with titanium abutments, (a) C1 implant at baseline with slight bone resorption, (b) circular C1 implants with less buccal bone resorption at 15 days, (c) buccal and lingual bone maintenance at 12 weeks in C1 implants, (d) circular implants with peek abutments at baseline, (e) C1 implant with peek abutment at 15 days with less bone resorption and (f) circular implant with peek abutment showing buccal and lingual bone maintenance at 12 weeks.

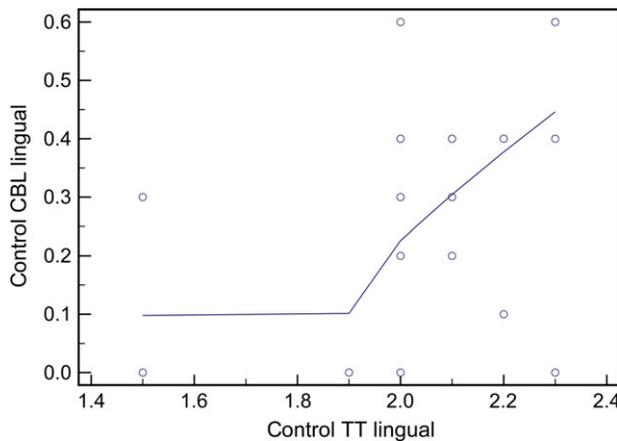


Fig. 10. The Pearson's correlations applied in the control group that showed a strong and statistically significant correlation between A-B and C-D parameters at the lingual site.

difference in the hole size and the implant configuration.

Meredith et al. (1997) demonstrated that the implants with higher initial ISQ values can reach the peak of the stability in shorter healing periods and concluded that such a method can serve as a useful research technique and may prove to be valuable in studying the behavior of implants in surrounding bone. In

addition, clinical observations indicated that the final healing time was affected by individual differences and operation conditions. During the process of osseointegration, the increasing rate of the ISQ values was approximately 300 Hz per week. Additionally, when the surgery was not successful, the ISQ values showed a 12% reduction during the first 2 weeks of healing (Meredith et al. 1997). In

the present study, all implants a higher ISQ values at baseline and 12 weeks after with no statistically significant differences. Applying the statistical test between the groups in the same time (baseline and 12 weeks), no statistically significant differences was found, so as within groups.

In the present study, the implants were positioned in the crestal bone level, following Bornstein et al. (2003, 2005) who reported that the implants are often inserted within the bone crest. Tomasi et al. (2010) in a clinical trial observed that the implant position conditioned the amount of buccal crest resorption. Moreover, the thickness of the buccal bone plate and the tridimensional positioning of the implant must be considered, because these are important factors that influence the response of hard tissues during healing.

Actually implants with change in platform (diameter of the abutment less to implant diameter) have demonstrated better crestal bone preservation. Then, this study was carried out by the insertion of implants with a change in platform and presented a SLA surface. Previous studies had established that the use of implants with a rough surface may influence the amount of bone regeneration and the values of BIC during healing (Abrahamsson et al. 2004; Botticelli et al. 2005; Abrahamsson & Cardaropoli 2007). Different studies have assessed that implants presenting a rough surface may influence the degree of bone regeneration and the percentages of BIC during healing (Wennerberg et al. 1996; Trisi et al. 2002; Botticelli et al. 2005). Calvo-Guirado et al. (2010, 2015) concluded that the surface treatment can reduce the crestal bone resorption. Cooper (2000) found that an increased surface roughness improves bone integration of the implant, increases osteoconduction, and increases osteogenesis.

New studies are needed to define the influence of others neck configurations on the crestal bone healing and the behavior with or without loading and the influence of abutment change on crestal bone stabilization. These would appear to be important factors for improving peri-implant bone and soft tissue stability and so clinical outcomes, including esthetics, which are of particular importance in the anterior zone. Our results suggest that more thickness of peri-implant tissues may be expected in this new implant design. However, the control group with traditional implant design was found to have more height values of the crestal bone because the bone is protected by a circumferencial design. Triangular head implants cannot be placed in hard bone because any blood supply can be expected.

Conclusions

Within the limitations of this study, our findings suggest that more thickness of peri-implant hard and soft tissues may be expected

in this new implant design. However, the control group with traditional implant design was found to have more height values of the crestal bone compared with new V3 implants.

Conflict of interest

The authors declare that they have no conflict of interest.

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