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Marginal bone and soft tissue behavior following platform switching abutment connection/disconnection– a dog model study

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Abstract

Objective: The effect on the marginal peri-implant tissues following repeated platform switching abutment removal and subsequent reconnection was studied.

Material and Methods: Six adult female Beagle dogs were selected, and Pm3 and Pm4 teeth, both left and right sides, were extracted and the sites healed for 3 months. At this time, 24 bone level (BL) (Straumann®, Basel, Switzerland) Ø 3.3/8 mm implants were placed, 2 in each side on Pm3 and Pm4 regions. In one side (control group), 12 bone level conical Ø 3.6 mm healing abutments and, on the other side (test group), 12 Narrow CrossFit™ (NC) multibase abutments (Straumann®, Basel, Switzerland) were connected at time of implant surgery. On test group, all prosthetic procedures were carried out direct to multibase abutment without disconnecting it, where in the control group, the multibase abutment was connected/disconnected five times (at 6/8/10/12/14 weeks) during prosthetic procedures. Twelve fixed metal bridges were delivered 14 weeks after implant placement. A cleaning/control appointment was scheduled 6 months after implant placement. The animals were sacrificed at 9 months of the study. Clinical parameters and peri-apical x-rays were registered in every visit. Histomorphometric analysis was carried out for the 24 implants. The distance from multibase abutment shoulder to the first bone implant contact (S-BIC) was defined as the primary histomorphometric parameter.

Results: Wilcoxon comparison paired test ($n = 6$) found no statistically significant differences (buccal $P = 0.917$; Lingual $P = 0.463$) between test and control groups both lingually and buccally for S-BIC distance. Only Pm3 buccal aBE-BC (distance from the apical end of the barrier epithelium to the first bone implant contact) ($P = 0.046$) parameter presented statistically significant differences between test and control groups. Control group presented 0.57 mm more recession than test group, being this difference statistically significant between the two groups ($P < 0.001$).

Conclusion: It can be conclude, within the limits of this animal study, that the connection/disconnection of platform switching abutments during prosthetic phase of implant treatment does not induce bone marginal absorption. Furthermore, it may present a negative influence in the buccal connective tissue attachment that becomes shorter anyway preventing marginal hard tissue resorption, especially in thin biotypes.

Multiple research groups have established that a biologic width (BW) exists around all dental implants (Hansson et al. 1983; Berglundh & Lindhe 1996; Hermann et al. 1997, 2001a). This is true for all implants of all shapes, whether on one-stage implants or after uncovering in two-stage placement protocols on two-piece implants (Berglundh & Lindhe 1996; Hermann et al. 1997, 2001a).

Peri-implant bone loss around implants exposed to the oral environment also has been documented extensively (Hansson et al.

1983; Berglundh & Lindhe 1996; Hermann et al. 1997, 2001a). Such resorption appears to be related primarily to exposure of the implant to the oral environment (Berglundh & Lindhe 1996; Baffone et al. 2013; Bengazi et al. 2013a,b). It has been demonstrated that the gap between the implant and the abutment has a direct effect on bone loss, regardless of whether the two parts are connected at the time of implant placement or after initial submergence and integration of the implant (Hermann et al. 1997). This phenomenon

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occurs whether the implant is loaded or unloaded and appears to be unrelated to the implant surface treatment (Cochran et al. 1997; Hermann et al. 1997). Management of such bone resorption is an important factor in achieving good esthetic results in the anterior maxilla and in optimizing bone support (Tarnow et al. 2000; Buser et al. 2004).

Vertical bone resorption, which often extends 1–2 mm below the implant–abutment interface, diminishes the bone-to-implant contact surface and, thus, impairs the biomechanics of restorations (Hermann et al. 1997, 2001b). Horizontal bone loss leads to resorption of the buccal plate in narrow alveolar crests, as well as loss of the interproximal bone peak and loss of support for the adjacent interimplant papilla. To reduce the effects of peri-implant bone resorption, a technique known as platform switching was recently developed (Lazzara & Porter 2006; Vela-Nebot et al. 2006; Calvo Guirado et al. 2007).

The concept behind platform switching is that by shifting the implant–abutment interface medially, the deleterious impact of the implant–abutment microgap on the peri-implant bone can be reduced. Platform switching thus involves the use of abutments with a diameter smaller than that of the implant platform (Lazzara & Porter 2006). This geometry shifts the perimeter of the implant–abutment junction inward toward the central axis of the implant (Baumgarten et al. 2005). It has been recently suggested that marginal bone level alterations could be related to the extent of implant/abutment mismatching being positively correlated with the amplitude of the mismatch (Canullo et al. 2010; Baffone et al. 2011, 2012). Marginal bone levels were better maintained at implants restored according to the platform switching concept (Canullo et al. 2010). While a reduction in horizontal bone resorption also has been observed in radiographs of platform-switched implants, the impact of platform switching on horizontal bone loss has not previously been directly studied and documented. Abrahamsson et al. (1997) further investigated the influence of the abutment dis/reconnection on the marginal peri-implant tissues (Bränemark System). The authors observed that abutment manipulation compromised the mucosal barrier and induced an apical migration of the connective tissue. Thus, while normal proportions and dimensions of the hard and soft tissues were observed in the control group, at test sites, the abutment manipulation resulted in a mechanical injury to the soft tissue barrier

that had to re-establish more apically, causing a marginal bone resorption (1.5 mm). In contrast, a single abutment reconnection proved to induce no marginal bone remodeling (Astra Tech Implant System) resulting in a transmucosal attachment of adequate quality and dimensions (Abrahamsson et al. 2003). More recently, Becker et al. (2012) in a dog model study found that 2 times (at 4 and 6 weeks) abutment dis/re-connection appeared to be associated with an obvious disruption of the established mucosal seal in the 12 switching platform implants investigated and concluded that repeated manipulation may be associated with dimensional changes of peri-implant soft and hard tissues formed at both mismatched Ti and ZrO₂ abutments.

The objective of the present experiment was to study histologically the effect on the marginal peri-implant tissues (hard and soft tissues) following repeated abutment removal and subsequent reconnection.

Material and methods

The experimental study was conducted during the year 2009 on a sample consisting of six Beagle dogs characterized for being castrated females and adults, with an average age of 23 months and an average weight of 15 kg. The handling of the animals was adjusted to Directive 86/609/EC on the approximation of laws, regulations and administrative provisions of the Member States regarding the protection of animals used for experimentation. Most of this policy is reflected in RD 1201/05 on November 21, 2005. Dogs in the study were provided by the Veterinary Faculty of Córdoba and installed in the service of animal experimentation hospital clinical veterinary “RoF Codina” in the Faculty of Veterinary of Lugo from the University of Santiago de Compostela. Care and maintenance of the animals took place on the campus of Lugo from the University of Santiago de Compostela, following the signs marked by the manuals for the care and use of laboratory animals from the phase of acclimatization until the time of sacrifice. The environmental conditions of temperature were $22 \pm 2^\circ\text{C}$ and a relative humidity between 50% and 70% on individual booths with 12: 12 h light/dark cycles. Food was with granulated feed throughout the experimental period, and the water was administered *ad libitum*.

The experimental area was located at the level of the Pm3 and Pm4 premolar region in each side of the mandible. Bone level (BL)

implants (Straumann®, Basel, Switzerland) made of pure type IV biocompatible titanium showing a rough SLActive® surface (Sand-blasted Large grit Acid etched) 3.3-mm diameter and 8-mm length were selected. Multibase Narrow CrossFit™ (NC) abutments (Straumann®) of 3.5-mm diameter, 1-mm height, made of pure type IV biocompatible titanium were selected (Fig. 1). These abutments were developed to screw retained multiple rehabilitations. Temporarily, during osseointegration time and prosthesis confection phase, there were used bone level conical Ø 3.6 mm healing abutments in control group implants and cover cups to the multibase abutments on test group implants.

Experimental design

Experimental procedure

Three months previously: teeth extraction

Teeth extractions were made in six beagle dogs in both sides of the Pm3 and Pm4 regions (Fig. 2). Surgical technique was similar in both control and experimental group. Teeth were carefully removed, separating the roots by means of tooth hemisection with the use of a fissure bur and extracting them individually with elevators and forceps. All the alveolus healed for a period of 3 months (0 months of the study).

Implant placement

After 3 months of healing (0 months of the study), implants were installed. Four BL Ø 3.3/8 mm SLActive® implants were placed (two in each side of the mandible – minimal 4 mm apart) by the same senior periodontologist. Implant shoulder was left at the level of cortical bone in all implants in both test and control groups. According to presurgical randomization, two multibase NC abutments were then screwed in one side (test group) of the mandible and two healing conical abutments were placed on the other side (control



Fig. 1. Multibase Narrow CrossFit™ abutments.

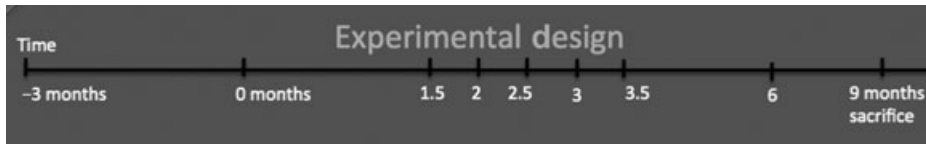


Fig. 2. Experimental design – time intervals in months.

group). Both multibase and healing abutments were screwed at 15 N (Fig. 3a–d). Post-operatively, pain was controlled with morphine (0.3 mg/kg/i.m.) during 24 h and meloxicam (0.1 mg/kg/s.i.d./p.o.; Metacam, Boehringer Ingelheim España, Barcelona, Spain) as analgesic during 3 days. Antibiotic prophylaxis was administrated during the first week with amoxicillin (22 mg/kg/s.i.d./s.c.; Amoxil retard, Syva, León, Spain). The dogs' diet throughout the trial period was granulated dog feed and had free access to drinking water. The animals were enrolled in a plaque control program consisting in cleaning the teeth and the implants three times a week with gauzes embedded in chlorhexidine oral rinse 0.12% during the first 2 weeks, and then a brush and toothpaste.

Impression taken

After 6 weeks of implant placement, the recommended period for SLActive® surface implants osseointegration, the healing screws were totally unscrewed and impression copings were screwed. Multibase abutments were screwed definitely at 35 N. Impressions were taken direct to implant on control side and direct to the abutment on test side.

Metal framework try-in

After 8 weeks of implant placement, the 12 test side multibase Narrow CrossFit™ abutments were not unscrewed and the metal prosthesis try-in was carried out. On control side, the healing abutments were removed, and 12 new multibase Narrow CrossFit™ abutments were connected for the try-in of the metal prosthesis. An ischemic marginal soft tissue was observed on control group, at the time of abutment connection as well as at the time of metal frame-work insertion (Fig. 4a,b), for opposition to the test group where no ischemic signal was clinically appreciated. Prosthesis adaptation was checked radiographically, and occlusal contacts were checked clinically for all 12 bridges. In one bridge, a premature contact was detected and corrected with a diamond bur. At 10 weeks of implant placement, a second metal framework try-in visit was scheduled and similar clinical procedures were carried out as in the first metal framework try-in. This appointment was designed to simulate clinical conditions were sometimes a second metal framework is necessary after mal-adaptation of the first one. At 12 weeks of implant placement, a bisc-bake prosthesis try-in was carried out repeating similar clinical

procedures on test and control sides. Prosthesis definitive delivery was carried out at 14 weeks. Healing abutments were removed on control side, and NC multibase abutments were placed again at this time and definitely screwed at 35 N. The bridges were definitively screwed at 15 N in each abutment and its perfect fit checked radiographically (Fig. 3c). Screws access holes were filled with silicone.

Cleaning control

After 6 months of implant placement, clinical parameters (bleeding index, plaque index, recession, suppuration, keratinized tissue, abutment mobility) were registered and all the bridges both on test and control sides were removed (not the multibase abutment) for cleaning control.

Dogs sacrifice

Nine months after implant placement and 6 months after prosthesis insertion, periapical X-Rays were taken of all six dogs and clinic parameters were again registered. Animals were sacrificed by an overdose of sodium pentobarbital (40–60 mg/kg/i.v.; Dolethal, Vetoquinol, France) previously sedated with medetomidine (30 µg/kg/i.m.; Esteve, Barcelona, Spain), and block section were taken and prepared for histology.

Clinical parameters

Plaque control was performed for every dog three times a week with a toothbrush and toothpaste. Clinical parameters were registered (bleeding index, plaque index, recession, suppuration, keratinized tissue, abutment mobility) at week 6/8/10/12/14 and 6 months. All clinical records were registered by the same blinded operator. Clinical records were taken in both groups according to the sequence described in the study of Buser et al. (1990).

Recession

Recession was measured taking into consideration the distance between implant shoulder (control group) or multibase abutment shoulder to the buccal marginal gingiva position, after removing Straumann® healing cups (control group) or Straumann® multibase Narrow CrossFit™ (NC) abutments cover protection (test group). This distance was measured with a manual millimeter periodontal probe Hu-Friedy® (Hu-Friedy, Chicago, IL, USA) on buccal surface.

Plaque and bleeding index

Plaque and bleeding index was measured with a manual millimeter periodontal probe

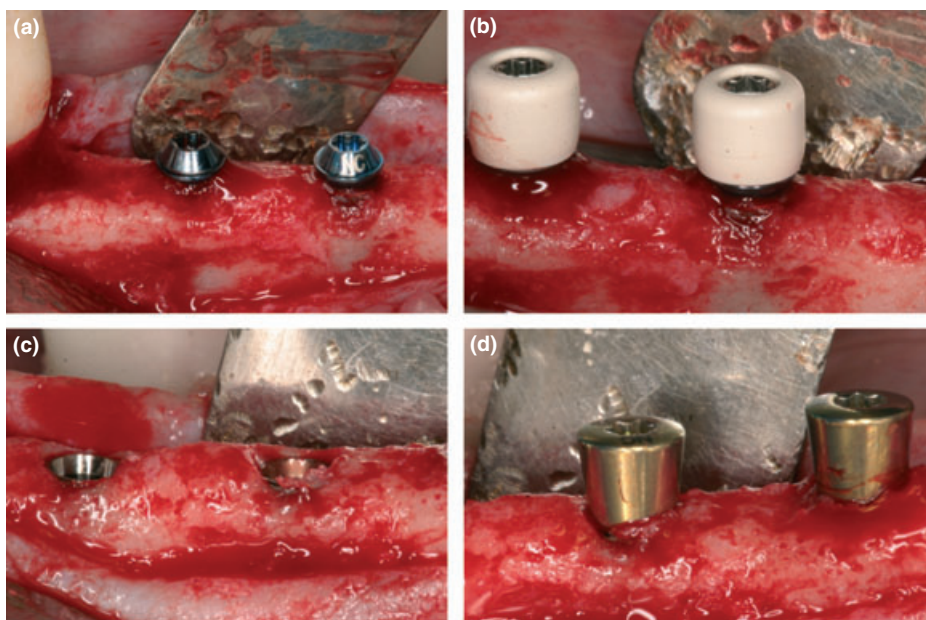


Fig. 3. [a–d] 2 multibase Narrow CrossFit™ abutments were screwed in test side of the mandible and two healing caps were placed on the control side of the six dogs.



Fig. 4. (a,b) Ischemic marginal soft tissue around implants was observed at the time that new multibase abutments were screwed and also at time of metal framework insertion, on group control. (c) X-ray control at time of definitive prosthesis delivery.

Hu – Friedy® (Hu-Friedy) on surfaces mesial, vestibular, distal and lingual of implants (Mombelli et al. 1987) passing it by the gingival sulcus surrounding the gingiva around implants.

Buccal keratinized gingiva height

Distance from buccal gingival margin to mucogingival line.

Suppuration

Inspection of clinical signs of suppuration exudate directly ascending from the peri-implant sulcus. Slight digital pressure from apical to coronal was applied on the marginal gingiva around implants both from test and control groups.

Abutment mobility

Clinical inspection of minor signs of Straumann® multibase Narrow CrossFit™ (NC) abutments mobility.

Radiographic parameters

Periapical digital x-rays were taken in each visit to both test and control group to check any sign of radiographic bone loss, loss of osseointegration, peri-implantitis or abutment/prosthetic unscrewing.

Histological processing

The lower jaw was removed and immersed in buffered formalin for 1 week. The four implants and the surrounding tissue were separated from each mandible using a diamond saw (Exakt; Apparatebau, Norderstedt, Germany). The biopsies were processed for ground sectioning in conformity with the method described by Donath and Breuner (1982). The samples were dehydrated using ascending grades of alcohol and embedded in a glycol methacrylate resin (Technovit 7200 VLC; Heraeus-Kulzer GmbH, Werheim, Germany). Sections of implants were glued to silanized glass slides and grinded to 40 µm. All

the slides were stained with Levai–Laczkó. From each implant, the most central buccal–lingual section was prepared for the histomorphometric analysis. Implants were histologically and histometrically analyzed using a light microscope (BX51; Olympus, Tokyo, Japan). By means of a color camera (DP71; Olympus), the images were captured and transferred to a computer. One calibrated masked examiner performed all the histomorphometric measures using a PC-based image analysis program (Microimage 4.0; Media Cybernetics, Silver Springs, MD, USA). The analysis was performed to evaluate the following variables on each group both buccally and lingually (Fig. 5). Linear measurements were made by drawing a

vertical line following the long axis of the implant as described:

- S-BC – Distance from the shoulder of multibase abutment to the bone crest.
- S-BIC – Distance from the shoulder of multibase abutment to the first bone implant contact.
- PM-BIC – Distance from the peri-implant margin to the first bone implant contact.
- PM-aBE – Distance from the peri-implant margin to the apical end of the barrier epithelium.
- aBE-BIC – Distance from the apical end of the barrier epithelium to the first bone implant contact, or rather the length in mm of the connective tissue of the peri-implant mucosa.

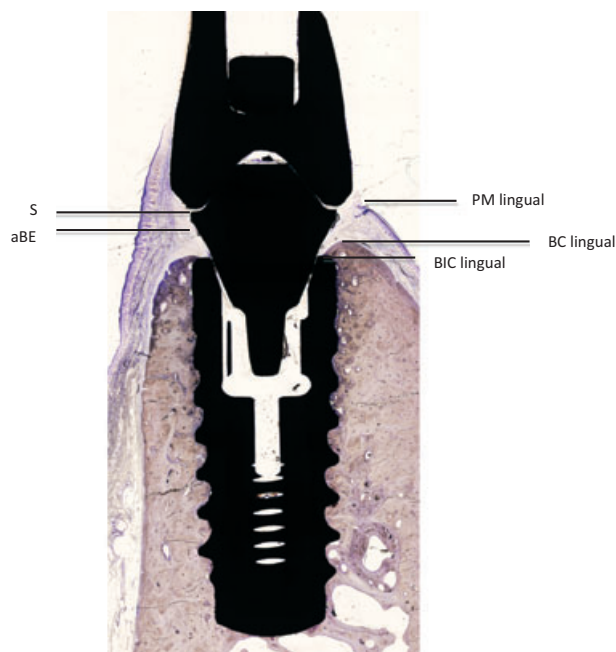


Fig. 5. Landmarks used for the histometrical measurements. PM, peri-implant mucosal margin; aBE, apical end of the barrier epithelium; S, multibase abutment shoulder; BIC, first bone-to-implant contact; BC, bone crest. Undecalcified ground B-L section, surface stained with Levai Laczkó – 1 mm magnification).

- S-PM – Distance from the shoulder of multibase abutment to the peri-implant mucosal margin.
- BC buc – BC ling – difference between buccal bone crest and lingual bone crest.

Statistical analysis

Average results across similarly treated implants in the same dog were calculated. Descriptive analysis was produced for each variable (mean values, standard deviation, median). Nonparametric Wilcoxon test was applied to evaluate histometric variables being S-BIC determined as the primary variable of this study. To evaluate clinical parameters of the study (recession and keratinized gingiva width), multivariate models of generalized estimation equations (GEE) were applied, with the identity as connection function, being assumed a linear evolution in time. This is a method that allows the examination of repeated or longitudinal measures, taking into account that the measurements in the same individual over time are correlated. The advantage of this method is that provides consistent estimated values of the parameters associated with covariates in the model. It was used a significance level of 0.05 for all tests of hypothesis. The analysis was carried out using the SPSS® v.19.0. statistical analysis program (IBM SPSS v.19.0. statistic program done at CIDES, Oporto Faculty of Medicine, Oporto, Portugal).

Results

Clinical observations

Twenty-four implants were placed in six dogs. One hundred percent implant and prosthesis survival were found at the end of the study. No adverse events occurred related to surgical or prosthetic protocol in any of the specimens, neither in test nor in control groups. No modification from the original experimental protocol was necessary. No health problems occurred to any of the animals until the time of sacrifice. Thus, all of the implants and all of the animals were available for analysis at the end of the study period. No clinical signs of suppuration neither radiographic signs of peri-implantitis were found at any of the scheduled visits of the study in both groups. At no visit, did the abutments (test group) or the healing abutments (control group) show any sign of mobility. At the 6 months cleaning control, none of the definitive prosthesis showed any sign of mobility. Applying multivariate mod-

els of generalized estimation equations (GEE) to clinical parameter such as Recession (Table 1) and Keratinized Gingiva (Table 2), there was found statistical significant differences between control and test groups only for parameter recession. There was found that control group (the group where the multibase abutments were not connect at time of implant placement – day 0) presented 0.57 mm more recession than the test group, which represents a statistically significant difference between the two groups ($P < 0.001$). No other clinical parameter analyzed on five consecutive visits (bleeding index, plaque index, suppuration and abutment mobility) showed any statistical significant difference between test and control groups.

Histological observations

Histological examination of the implant samples revealed that the peri-implant mucosa was covered by a keratinized oral epithelium, which in the marginal border connected with a thin barrier epithelium of a few cell layers thickness and an underlying organized connective tissue. Similar observations were found for test and control groups (Fig. 6a,b).

Histometric results

Mean values found for buccal S-BIC (test group = 0.53 mm; control group = 0.52 mm) and lingual S-BIC (test group = 0.13 mm; control group = 0.11 mm) were very similar for both groups. Also mean values found for buccal S-BC (test group = 0.48 mm; control group = 0.42 mm) and lingual S-BC (test group = 0.16 mm; control group = 0.11 mm) were very similar for both groups. The difference between buccal BC and lingual BC presented a mean value of 0.60 mm for test group and 0.54 mm for control group. Being defined the histometric parameter S-BIC

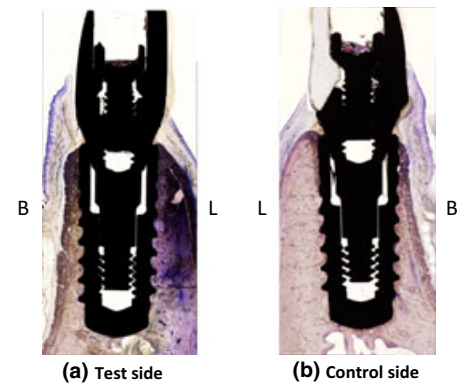


Fig. 6. (a,b) Histological preparation of test (a) and control (b) side implants (B-buccal, L-lingual). Undecalcified ground sections, surface stained with Levai Laczkó – 1 mm magnification.

(distance from the shoulder of multibase abutment to the first bone implant contact) as the primary variable of the study, nonparametric Wilcoxon comparison paired test ($n = 6$) found no statistically significant difference (buccal $P = 0.917$; Lingual $P = 0.463$) between test and control groups both lingually and buccally (Table 1). There were also found no statically differences for S-BC (buccal $P = 0.600$; lingual $P = 0.345$) and for buccal BC–lingual BC ($P = 0.48$) between test and control groups (Table 3). Mean values for buccal PM-BIC (test group = 3.80 mm; control group = 3.88 mm), buccal PM-aBE (test group = 2.79 mm; control group = 3.13 mm), buccal aBE-BC (test group = 1.10 mm; control group = 0.65 mm) and buccal S-PM (test group = 3.27 mm; control group = 3.36 mm) were found. On lingual side, mean values for parameters lingual PM-BIC (test group = 2.03 mm; control group = 1.98 mm), lingual PM-aBE (test group = 1.26 mm; control group = 1.35 mm), lingual aBE-BC (test group = 0.39 mm; control group = 0.39 mm)

Table 1. GEE model for clinical parameter recession (mm)

	Beta	IC 95%	P
Multibase (day 0)			
Yes	–	–	–
No	0.577	0.330	0.825
Constant	0.344	–	–

Table 2. GEE model for clinical parameter keratinized gingiva (mm)

	Beta	IC 95%	P
Multibase (day 0)			
Yes	–	–	–
No	0.113	–0.166	0.392
Constant	0.724	–	–

Table 3. Comparison paired test for test and control groups for S-BIC; S-BC and BC Buc – BC Ling (Buccal and Lingual) ($n = 6$) (mm)

	Mean	SD	Median	P
Lingual – S-BC				
Test	0.16	0.10	0.16	0.345
Control	0.11	0.34	0.15	
Lingual – S-BIC				
Test	0.13	0.08	0.14	0.463
Control	0.11	0.26	0.02	
Buccal – S-BC				
Test	0.48	0.21	0.39	0.600
Control	0.42	0.30	0.53	
Buccal – S-BIC				
Test	0.53	0.20	0.52	0.917
Control	0.52	0.29	0.57	
BC Buc – BC Ling				
Test	0.60	0.33	0.68	0.48
Control	0.54	0.27	0.54	

and lingual S-PM (test group = 1.76 mm; control group = 1.86 mm) were found. Wilcoxon test for histometric parameters found also no statistically differences between test and control groups (Table 4). Applying Wilcoxon comparison paired test to Pm3 and Pm4 implants separately, only for Pm3 the parameter buccal aBE-BC ($P = 0.046$) was found statistically different between test and control groups (Table 5).

Discussion

The dimensions of biologic width between teeth and implants are similar being approximately 2 mm of epithelial tissue and 1–1.5 mm of connective tissue (Berglundh et al. 1991; Abrahamsson et al. 1996). A minimum dimension of the biologic width is needed to accommodate for the soft tissue healing process: when this dimension is not present, bone resorption may occur, to allow for an “appropriate biologic dimension” of the peri-implant soft tissue barrier. The biologic width determines the minimum

dimensions of peri-implant mucosa that ensure adequate junctional epithelium and supra-alveolar connective tissue to maintain an optimal seal around implants and provide protection from mechanical and external biologic agents (Linkevicius et al. 2009, 2010). When an external agent invades the biologic width, the epithelium responds by migrating beyond the damaging agent in an attempt to isolate it and create a defensive distance that ensures periodontal integrity. This results in bone resorption, which ensures the reestablishment of the biologic width dimensions. This process is also observed around natural teeth when the biologic width is invaded by formation of calculus or infra-gingival margins of crowns.

The establishment of biologic width can be affected by the surgical technique, loading, microgap, implant position, infection/inflammation, switching platform concept, immediate implants flap vs. flapless (Blanco et al. 2008) and abutment manipulation. Microgap between implant and abutment, when present, can modify the dimension of biologic width. The longer epithelial component described may be determined by bacterial colonization or abutments micromovements. Hermann et al. (2001a) has reported that the bone loss at the alveolar crest is significantly influenced by micromovements of the implant components, but not by the size of the microgap. They concluded that significant crestal bone loss occurs in 2-piece implant configurations, even with the smallest-sized microgaps ($<10\ \mu\text{m}$) in combination with possible movements between implant components. The width of the interface, micromovements of the implant and/or abutment and peri-implant vascular alterations might all contribute to the influence of microbial contamination on the biologic width (King et al. 2002). It was suggested that healing abutment disconnection as a part of prosthetic treatment results in disruption of the epithelial seal, causing bleeding and ulceration of the site. This mechanical disruption may be considered as an open wound or exposure of connective tissue, which may result in inflammatory responses and epithelial migration. The re-establishment of biologic width in a more apical position may be one of the factors that could explain initial crestal bone loss. Although each and every of these factors may contribute to the establishment of biologic width, in this particular animal study, we have tried just to vary the factor abutment manipulation and analyze its possible impact on the behavior of biologic width.

In our study, we have found no statistically significant differences concerning the distance between implant shoulder of multibase abutment to the first bone implant contact (S-BIC) when there was or not abutment manipulation, what could mean that, at least for switching platform implants, five time abutment connection/disconnection during prosthetic phase seem not have influence on marginal bone stability. We have found statistically significant differences for Pm3 concerning the parameter buccal aBE-BC ($P = 0.046$). The distance from the apical end of the barrier epithelium to the first bone implant contact or rather the length in mm of the connective tissue of the peri-implant mucosa at the buccal side (buccal aBE-BC) in the test group was longer than in control group (abutment manipulation). This could mean that the connective tissue length in control group would become shorter in time probably do to the disruption of connective tissue attachment at time of abutment manipulation. Although the connective tissue length appears to be shorter after abutment manipulation, it would still prevent the apical resorption of buccal crestal bone, as the S-BIC distance did not significantly varied. The reason that may justify why the statistic significant differences were found on the connective tissue component of the biologic width and not on the epithelial component could be related with the fact that with the NC abutments, the platform switching concept is applied. So, the apical reorganization of the biologic width components due to abutment manipulation could be carried out mainly in response to connective tissue horizontal and vertical changes than to the apical crestal bone resorption or to epithelial attachment changes. It seems that the epithelial part of biologic width after platform switching concept abutment manipulation would reattach, maintaining its length, and that the connective tissue component would reorganize, becoming shorter. It seems that platform abutment manipulation plays an influence on the connective tissue portion of implant biologic width, which becomes shorter but does not conduct to more buccal marginal bone resorption, protecting the bone to reabsorb. This seems especially true for the buccal soft tissue margin around Pm3 where keratinized tissue is thinner than Pm4 region. Becker et al. (2012) in a dog model study found that two times (at 4 and 6 weeks) abutment dis/re-connection appeared to be associated with an obvious disruption of the established mucosal seal in the 12 switching platform implants

Table 4. Comparison paired test for test and control groups for PM-BIC, PM-aBE, aBE-BIC and S-PM (Buccal and Lingual) ($n = 6$) (mm)

	Mean	SD	Median	<i>P</i>
Lingual – PM-BIC				
Test	2.03	0.34	1.89	0.463
Control	1.98	0.38	1.99	
Lingual – PM-aBE				
Test	1.26	0.25	1.19	0.600
Control	1.35	0.57	1.50	
Lingual – aBE-BC				
Test	0.39	0.31	0.29	0.917
Control	0.39	0.41	0.28	
Lingual – S-PM				
Test	1.76	0.55	1.80	0.249
Control	1.86	0.46	1.84	
Buccal – PM-BIC				
Test	3.80	0.57	3.83	0.463
Control	3.88	0.57	4.06	
Buccal – PM-aBE				
Test	2.79	0.56	3.06	0.249
Control	3.13	0.72	3.09	
Buccal – aBE-BC				
Test	1.10	0.31	1.03	0.249
Control	0.65	0.46	0.79	
Buccal – S-PM				
Test	3.27	0.53	3.47	0.600
Control	3.36	0.52	3.47	

Table 5. Histological data: paired comparison between test and control sides for Pm3 ($n = 6$) (mm)

	Mean	SD	Median	<i>P</i>
Buccal aBE-BC				
Test	0.95	0.34	0.99	0.046
Control	0.53	0.30	0.55	

investigated and concluded that repeated manipulation may be associated with dimensional changes of peri-implant soft and hard tissues formed at both mismatched Ti and ZrO₂ abutments. Anyway these conclusions were based on the results of only three dogs (12 implants) which histometrical analysis was carried out at 8 weeks, meaning that the soft tissue only had 2 weeks between the last abutment dis/re-connection (6 weeks) until dog's sacrifice to heal and eventually reorganize. In our study, apart from the 5 times abutment dis/re-connection, the soft and hard tissue was left undisturbed for a period of almost 6 months (extrapolating 18 months in humans) between the last abutment dis/re-connection and dog's sacrifice to ensure sufficient soft and hard tissue stabilization. Berglundh and Lindhe (1996) reported that thin tissues may provoke crestal bone loss, during the formation of the peri-implant seal in an animal experiment. Observations in other histological study showed that implants, surrounded by consistently thin mucosa, had angular bone defects, while at implant sites with even alveolar pattern, wide mucosa biotype was prevalent (Abrahamsson et al. 1996). The histometrical analysis of this study failed to corroborate these facts. On the other hand, the clinical results for buccal Recession found statistically significant differences ($P < 0.001$) between test and control groups showing that the control group presented 0.57 mm more recession than the test group. More Recession may be the clinical result of the immediate disruption of the connective and epithelial biologic width attachment at time of abutment manipulation in each appointment at control group. The clinical recession parameter was measured immediately after healing abutment disconnection and multi-base connection. This was special significant on Pm3 region. These findings may indicate that the connection/disconnection of the abutment during the prosthetic phase of implant treatment may present an immediate influence on the buccal soft components of biologic width, especially in thin biotypes, and that a shorter horizontal connective attachment component at switching implant shoulder will reorganize to prevent bone marginal bone resorption. This is corroborating with the clinical observation of an ischemic marginal implant soft tissue present on control group at every time that the healing abutments were disconnected and the NC abutments were connected for successive prosthesis try-in visits (five visits), in opposition to the test group where no ischemic

signal were clinically appreciated. These findings should be considered mainly in esthetic regions where some uncontrolled soft tissue recession during prosthetic phase may compromise final rehabilitation. Blanco et al. (2010) also found in a dog model study that the mean values for the biologic width longitudinal dimension at the buccal aspect were higher in the flap group than in the flapless group on immediate implants and this difference mostly being due to the Pm3, probably because of a thinner biotype in this region. It is well acknowledged that stability of crestal bone around implants plays a major role in implant longevity and esthetic outcome of treatment. A stable bone level around the implant neck is a prerequisite for achieving support and, hence, long-term optimal and stable gingival contours. This is especially so with regard to the interdental papillae in the anterior region. Thus, even loss of 0.5 mm may result in poor esthetics or dramatically disturbed bone-to-implant contact of a short implant. The ability to predict the amount of bone remodeling around implants is important for a stable and predictable esthetic result. The purpose of the Hartman and Cochran (2004) study was to investigate the amount of radiographic bone remodeling that occurs over time using a one-piece implant system. They selected 27 patients receiving implants in the maxilla, and 15 receiving implants in the mandible were included in the study. All implants were placed with a non-submerged surgical technique with varying locations of the rough-smooth border with respect to the alveolar crest. Clinical exams and radiographs were taken on the day of implant placement, at 6 months, and annually up to 5 years. They found that a significant amount of bone remodeling compared to baseline occurred for all implants at the 6-month follow-up visit (1.10 mm), with the remaining time points showing virtually no change (0.1 mm). A relationship was found between the amount of bone remodeling and the location of the rough-smooth border with respect to the alveolar crest. Those implants with the rough-smooth border surgically placed below the crest had, on average, a greater amount of remodeling at 6 months (average 1.72 mm) than implants with the rough-smooth border placed at or near the crest (average = 0.68 mm). In both situations, this remodeling (i) occurred early (within 6 months), (ii) reached a similar level and (iii) remained virtually unchanged up through 60 months (0.05 mm). These results lead to the conclusion that a physiologic dimension appears to exist between the bone

and the implant-crown interface around one-piece implants that is established early and maintained over time. These results are significant because they demonstrate in patients that the magnitude of initial bone remodeling around these one-piece dental implants is dependent on the positioning of the rough-smooth border of the implant in an apico-coronal dimension. In this study, the 24 BL implants were placed at bone crestal level, both in test and control groups to avoid bias concerning initial biologic width remodeling of implants placed below bone crestal level. In this study, the rough SLActive implant surface and its insertion position (at bone crest level) may also have contributed to bone level crest stability outcomes that were found. Degidi et al. (2010) observed from an human study that a small but significant horizontal bone loss (non vertical) was evidenced in the hard tissue portion over the subcrestal implant platform in a period of time between the 6-months and 1-year follow-up when the abutment was removed to impression taken compared with the "one abutment at one time" concept when the abutment was applied immediately at time of implant placement. This difference seemed to be maintained over the 3-year period of the study. Grandi et al. (2012) found for 28 patients with partial edentulism rehabilitated with two implant supported immediate restoration that when definitive platform-switched abutment was connect at time of surgery, there was statistically less bone resorption than in the group where abutments were removed for impression taken. Thus, many studies presently aim and surely will focus in the future on determination of the clinical and technical solutions, which must be undertaken to prevent recession of soft and hard tissues. Abutment time of connection/manipulation will always be a factor to be considered by the clinician and its impact in the overall treatment success is of utmost importance. There is a lack of research data regarding abutment time of disconnection/re-connection influence on stability of crestal bone around implants (Rompen 2012). It appears that almost no clinical studies have been found in the literature on the topic and very few animal experiments evaluated this relationship. However, in light of evidence-based dentistry, results from animal studies cannot be directly attributed to clinical practice, and they play a definitive role for the understanding of the processes to elaborate clinical trial. The influence of dental implants on the surrounding soft and hard tissue is crucial in defining the implant's

functional and esthetics success. Therefore, more histological and clinical research will be necessary to confirm or reject these animal results.

Conclusion

It can be concluded, within the limits of this animal study, that the connection/disconnection

of platform switching abutments during prosthetic phase of implant treatment does not induce bone marginal reabsorption. Furthermore, it may present a negative influence in the buccal connective tissue attachment that becomes shorter anyway preventing marginal hard tissue resorption, especially in thin biotypes. These findings should be considered especially in esthetic regions where some

uncontrolled soft tissue recession during prosthetic phase may compromise final rehabilitation.

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