



A Comparison of Zirconia CAD/CAM to Conventionally Fabricated Single Implant Restorations in the Esthetic Zone

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A Comparison of Zirconia CAD/CAM to Conventionally

Fabricated Single Implant Restorations in the Esthetic Zone

A Thesis Presented by

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То

The Faculty of Medicine

In partial fulfillment of the requirements for the degree of

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Dedication

I dedicate my DMSc Thesis work to my parents for their endless love, support, and encouragement and for everything they have done to make who I am today.

To my wife Azzah, who encircled me with her love, support and encouragement during my DMSc journey and life. Also, I dedicate this work to my daughter Layal who brought happiness to our family since her birth.

To my sisters, brothers, Father-in-law and Mother-in-law, who have always loved and supported me.

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Abstract

Objective: This project aimed to determine whether single tooth implant restorations fabricated with CAD/CAM zirconia abutments/porcelain fused to zirconia crowns reveal different biological and esthetic outcomes compared with prefabricated anatomic titanium abutments/porcelain fused to metal crowns in the esthetic zone.

Materials and Methods: Thirty patients who needed a single implant restoration in the esthetic zone were included in the study. Twenty-nine patients completed screening, baseline, crown insertion, one-month, six-month and one-year followup visits. At the screening visit, the patients were randomly allocated into two groups: the prefabricated anatomic titanium abutments/ porcelain fused to metal crown (Ti) group and the CAD/CAM zirconia abutments/ porcelain fused to zirconia crowns (Zr) group. Plague and bleeding scores, microbial profiles, probing depth, width of keratinized tissue, vertical bone changes, pink and white esthetic scores, papilla height, and clinical crown height were evaluated through several study time points. Furthermore, patients' self-esteem, satisfaction, and esthetic evaluations were assessed using visual analog scores (VAS). A simple scale called subjective and objective esthetic classification (SOE) was created to assess the esthetic success of treatment. Statistical analyses were performed using the Mann Whitney U test, Chi-square test and a generalized linear mixed model.

Results: All implants were successfully osseointegrated with a 100% survival rate over one year. Biologically, both groups had comparable outcomes except that the mean prevalence of the bacteria in the Zr group was significantly greater than in the Ti group at the final visits for *Streptococcus intermedius* (p< 0.0001). Also, the *Treponema denticola* DNA probe signal was lower in the Zr group than the in Ti group at the final visits (p= 0.0007). In addition, the mean of probing depth of the mesial tooth at the mesio-lingual site (p= 0.02) was less in the Zr group. All the esthetic parameters showed no statistically significant differences between both groups. Patients' self-esteem, satisfaction, and esthetic evaluations did not differ between groups.

Conclusion: After one year of clinical performance, the Zr group showed comparable results to the Ti group. This indicated that good clinical, biological and esthetic outcomes could be achieved by either treatment option. Further observations and follow-up are required to evaluate long-term results.

Background

Several long-term prospective studies have demonstrated the high success rate for dental implants with early and conventional loading protocols [1-5]. However, the longevity of dental implants depends on maintaining the health of peri-implant tissues and avoiding bacterial infection [6]. Before 1990, the most commonly used approach for restoring single dental implants was either prefabricated or custom abutments and porcelain fuse to metal crowns [7]. Although this conventional treatment approach has achieved acceptable results, several disadvantages have been reported. The presence of microscopic gaps between the different components, surface roughness of abutments and prostheses, and over-contouring of implant prostheses facilitated bacterial colonization and plague collection [8-10]. Moreover, surface roughness of the abutment and prosthetic materials can enhance biofilm formation. For instance, Quirynen et al. found that biofilm formation around roughened Ti abutments is 25 times greater than the standard Ti abutments after three months evaluated using DNA probe analysis and cell culturing techniques [11]. Infection and associated inflammation from bacterial colonization can significantly affect the integrity of oral Ti implants and the prostheses [12, 13]. The metallic color of the Ti abutments can cause gray to bluish discoloration of the overlying mucosal tissue, which, in turn, affects mucogingival esthetics in implant-supported prostheses [14]. Furthermore, the predetermined shape of the prefabricated Ti abutments

compromises the establishment of an optimal emergence profile [7], which may reduce the esthetics outcome.

Computer-aided design and computer-aided manufacturing (CAD/CAM) technology was introduced to implant dentistry to help improve the quality of abutments and prostheses [7, 15]. This technology allows the fabrication of implant abutment and/or prosthesis from a solid block of material that has a homogeneous composition and high mechanical properties [15]. Several laboratory steps, such as waxing, investing, or casting, are eliminated, resulting in the production of abutment and/or prosthesis with high quality and precision [7, 15]. Moreover, an appropriate emergence profile can be easily designed and achieved [15]. Furthermore, the visual properties of CAD/CAM ceramic abutments are closer to those of the natural tooth, and they may provide an appropriate esthetic outcome for the adjacent soft tissues [15, 16]. The use of CAD/CAM technology in the production of implant ceramic frameworks and crowns, especially in the esthetic zone, has been expanding rapidly because of the esthetic, biological, and mechanical benefits of the resultant ceramic materials.

In general, ceramic abutments are characterized by good soft tissue biocompatibility [16-18]. In addition, they maintain the marginal bone around a single-tooth implant at levels comparable to that achieved by abutments made of Ti [19]. In a prospective clinical study, plaque accumulation around dental implants with ceramic-coated transmucosal elements was significantly reduced

compared with plaque accumulation around Ti abutments [20]. These results suggest that further development of ceramic materials in implant dentistry may assist in plaque control and, thus, favorably influence the health of the soft tissues [20].

Zr oxide ceramics were introduced into implant dentistry to take advantage of their beneficial properties. Current studies on soft tissue responses to Zr abutment indicate good biological outcomes. Welander et al. found that soft tissue healing around Zr abutments was similar to the tissue around Ti abutment after five months when tested in Labrador dogs [21]. Degidi et al. performed immunohistochemical analysis on soft tissues harvested from five patients who had received dental implants with healing caps made of Zr and Ti to assess the peri-implant soft tissue responses [22]. They observed that mucosal tissues surrounding Zr healing caps expressed lower levels of inflammatory responses than the tissues surrounding the Ti caps after 6 months of healing [22]. Few studies, however, have observed that Zr abutments were surrounded by healthy soft tissues are clinically comparable to those around Ti abutments [23-25]. Radiographically, a study by Ekfeldt et al. demonstrated minimal bone loss around the Zr abutments, and they concluded that Zr abutments for single implant restorations have good biological results [25]. Other studies reported comparable amount of vertical bone loss between the Zr and Ti abutments [16, 23, 24].

Inflammation of the peri-implant mucosal tissues due to plaque accumulation may advance to bacterial infection and, eventually, to periimplantitis [26]. Although the use of all-ceramic abutments may reduce the plaque adherence around the gingival sulcus of the implant [26], limited numbers of studies have investigated the bacterial colonization on Zr oxide [27-31]. Scarano et al. found that Zr disks were characterized by less bacterial adhesion determined from scanning electron microscopy (SEM) than pure Ti disks after 24 hours of exposure to the oral cavity [29]. Likewise, Rimondini et al. found that, after three months, Zr disks had accumulated less bacterial load than commercially pure Ti [28]. Moreover, SEM evaluation for Zr disks in a split-mouth study design indicated that the polished and glazed Zr disks had similar bacterial colonization with less plaque biofilm on the polished surfaces [30]. Furthermore, after 4 hours, Zr disks placed on the buccal side of a removable partial denture had a lower concentration of bacteria than polycrystal alumina and hydroxyapatite disks [27]. All these studies highlight the potential advantages of using Zr oxide to restore dental implants. However, these reports used Zr disks splinted to removable devices, which does not reflect the actual clinical situation where the abutments are surrounded by the biological tissues with associated supragingival and subgingival microbiotas. These microbiotas are typically composed of different types of bacterial species [32, 33], and their adherence to Zr abutment needs further examination. Microbiology assessments have been

limited to SEM observations, which differentiates cell shape but not bacterial species identity [27-30].

Few studies have examined the oral microbiota, comprising about 700 bacterial species, of Zr compared with Ti implants. PCR based studies targeted only 2 or 7 periodontally pathogens around the Zr abutment [34-36]. These clinical studies did not show any differences between the Zr and Ti abutments in the adherence of selected periodontally pathogenic bacteria [34-36]. The follow-up period of these studies ranged from two weeks to three months, which was too short a time to evaluate material aging and fatigue over time. Furthermore, sufficient information about the most common oral bacterial species that may adhere to Zr is lacking and needs further investigation in partially edentulous patients. The DNA-DNA checkerboard hybridization approach has been used to detect up to 40 commensal and pathogenic bacteria [33, 37, 38], and can provide information to understand the nature of microorganisms that adhere to Zr abutments.

Maintaining the integrity of the hard and soft tissues around dental implants and abutments improves the esthetic outcome of the tooth replacement therapy. Guidelines for fixed implant restorations for optimal esthetics involve having healthy peri-implant tissues and prosthetic restorations in harmony with surrounding dentition [39]. A few recent studies have shown positive esthetic outcomes with Zr abutments surrounded by healthy peri-implant mucosa using spectrophotometry [24, 40, 41]. Bressan et al. found that the use of CAD/CAM Ti

abutments in the anterior maxilla altered the peri-implant soft tissue color when compared to CAD/CAM Zr abutments, as detected by spectrophotometric analysis [40]. Moreover, results from another study indicated that the placement of Zr abutments in the anterior maxilla tended to provide better esthetic outcomes than with Ti abutments [42]. However, Zr abutments had affected the color of the soft tissue around the dental implant when compared with the gingival color around natural teeth [24]. This change in the color was minimal when compared to other metallic abutments [43]. Further, the use of spectrophotometry provides information about the color of the selected area only. Therefore, the use of pink and white esthetic scores (PES and WES) as proposed in the current study provides a comprehensive evaluation of 10 esthetic parameters from an objective clinical perspective, as described by Belser et al. [44]. Only two previous studies have used the PES and WES scores to analyze the esthetic outcomes of Zr abutments and crowns [45, 46]. The first study was a case report for a singletooth implant restored with a Zr abutment and crown [45]. The second study did not account for changes that may occur at different time points in the soft tissue from baseline to the one-year follow-up [46]. Another limitation of the Furze et al. study was the small sample size of 10 patients [46]. Therefore, further studies are indicated to augment the esthetic outcome measures in clinical studies.

Soft tissue redness from gingival inflammation following bacterial accumulation or grayish discoloration due to the use of Ti abutments can affect patients' satisfaction with the overall appearance. Patients' satisfaction with the

treatment outcome is increasingly being considered to be a key consideration for treatment success [47]. The number of studies incorporating patients' centered outcomes is still limited in the field of implant dentistry, particularly those examining the impact of single-implant restorations on the quality of life. Only two studies have investigated whether different abutments for single implant restorations can affect patients' quality of life. These studies have reported satisfactory results from the overall treatment esthetic outcome when metallic and Zr abutments were used to restore single dental implants [48, 49]. However, the satisfaction level of the esthetic outcome can differ between patients and clinicians as each one of them may evaluate the esthetic according to different standards [44, 50, 51]. Knowing the appropriate treatment options that increase patients' satisfaction would help clinicians provide better care to their patients. Hence, there is a necessity to create a new scale that incorporates clinician and patient's esthetic assessments. This scale would provide the clinicians with additional treatment information related to the esthetics of the final crown before its delivery, and improve patients' awareness regarding the standards of esthetics.

Another important aspect of quality of life is patients' self-esteem. Losing a tooth in the esthetic zone, gingival display with grayish discoloration, or mouth odor due to plaque and bacterial buildup usually affects patients psychosocially, which, in turn, may reduce their self-esteem. A recent qualitative study by Atieh et al. evaluated patients' self-esteem when they were treated with immediate

single molar implants [52]. They found single molar implants did not affect the self-esteem [52]. This may be because molar teeth are not visible to others, and teeth esthetic usually does not rely on posterior teeth. Investigations are needed, however, to assess the effect of replacing an anterior tooth with different abutments materials on patients' self-esteem, satisfaction and report their esthetic evaluations.

Unfortunately, previous ceramic abutments on implants have exhibited more sensitivity to fracture than Ti abutments [19]. However, advances in the fabrication of high-strength all-ceramic abutments allowed their use to restore anterior implants in the esthetic zone [53]. The hardness of the Zr ceramic allows it to withstand conventional prophylaxis with ultrasonic scalers without altering the surface quality of the abutment [54]. Moreover, Zr has flexural strength and resistance to fracture almost twice as high as alumina [16, 53]. CAD/CAM Zr abutments are milled from solid homogeneous blocks with high mechanical properties [15], although it is still recommended that the thickness of the Zr is kept above 0.5 mm to avoid fracture [55]. Recently, Zr abutments in the posterior regions demonstrated good survival without fracture up to five years of follow-up [23, 56]. Complications have been reported in other studies when Zr abutments were used including abutment fracture, abutment screw loosing and veneer porcelain chipping [42, 48, 56]. Therefore, documenting the biological, mechanical, and technical complication rates of CAD/CAM Zr abutments and

prosthesis could add to current evidence to support the use of this material in implant dentistry.

The proposed study is specifically intended to provide data for bacterial colonization, peri-implant soft tissue parameters, and vertical bone changes for single implants in the esthetic zone restored with CAD/CAM Zr abutment/ porcelain fused to Zr crowns in comparison to prefabricated anatomic Ti abutments/porcelain fused to metal crowns in partially edentulous patients; the study design also is intended to account for material aging over time by using specific time points. The study will additionally provide evidence about esthetic outcomes, patients' satisfaction, and complication rates of the implant-supported prosthesis in the two tested groups. Finally, this study will provide a new scale that facilitates clinical decision-making as it relates to the choice of the final prosthesis and adjacent tissue esthetic outcome.

Specific Aims

Several features have indicated potential advantages of CAD/CAM Zr abutments over Ti abutments. First, less bacterial colonization was found on Zr than on Ti disks [28-30]. These results may indicate improvement in peri-implant soft tissue health and inhibition of gingival inflammation and bone loss when Zr material is used to fabricate implant abutments and crowns. Second, CAD/CAM implant abutments constructed with Zr may have better esthetic outcomes when compared with prefabricated anatomic Ti abutments. An *in vitro* investigation indicated that Zr resulted in minimal mucosal color changes when compared to Ti [43]. Since the metallic discoloration from Ti can be avoided, patients' satisfaction based on esthetic outcome may increase. Finally, the fabrication of CAD/CAM Zr abutments requires fewer laboratory steps that may, in turn, minimize the incidence of technical errors [15]. However, this rationale calls for a comprehensive comparative evaluation to test all these variables in a clinical setting, under investigation, and in a selected cohort of patients.

Hypothesis

There are differences in the peri-implant soft tissue parameters, bacterial colonization, vertical bone changes, esthetics outcomes, and patients' centered outcomes between the Zr and Ti abutment groups.

To test this hypothesis, the following four aims are proposed:

Specific Aim 1: To compare the biological response by means of bacterial colonization, peri-implant soft tissue parameters, and vertical bone changes in the esthetic zone between Zr and Ti groups.

1.1 Analysis of bacterial colonization: Plaque samples from the deepest area of the adjacent tooth sulcus and the peri-implant sulcus will be collected to detect the prevalence and relative proportions of 40 commensal and pathogenic bacteria using the DNA-DNA checkerboard hybridization approach.

1.2 Evaluation of peri-implant soft tissue involves several measures over time. Measurements include adjacent teeth and implant-prosthetic complex plaque and bleeding scores and the width of facial keratinized mucosa. Probing depth around the adjacent teeth will be measured too.

1.3 Measurement of the vertical bone changes (mesially and distally) around Zr and Ti groups by using consecutive radiographic evaluations with a specific reference landmark.

Specific Aim 2: To compare the esthetic outcomes between the Zr and Ti groups using objective parameters.

2.1 Esthetic outcomes will be evaluated by measuring the PES and WES as performed by a trained clinician at each visit. The sum of the total PES and the total WES will represent the total esthetic score (PES/WES).

2.2 Diagnostic casts of the implant-prosthetic complex and the adjacent teeth will be used to measure mesial and distal papilla heights and the clinical crown heights of the adjacent teeth and the implant crown.

Specific Aim 3: To compare the extent of patients' satisfaction with the esthetic outcomes between the Zr and Ti groups over time.

3.1 Outcome variables of patients' overall treatment satisfaction, self-esteem after replacing a missing tooth, and esthetic evaluation of the treatment provided will be measured. The difference between patients' expectations and final treatment outcomes will be evaluated using a visual analog scale.

3.2 PES/WES scores and the patients' esthetic scores will be used to develop a new scale to assess success of treatment esthetic outcomes. Based on the obtained percentages, the final prosthesis and the condition of adjacent soft tissues will be classified into three categories: satisfactory, marginal, or unsatisfactory.

Specific Aim 4: To report any mechanical, biological, and technical complications arising from the implant and/or the prosthesis in the esthetic zone between the Zr and Ti groups in partially edentulous patients.

Innovation and Clinical Significance

1. This study, for the first time, assesses the prevalence and proportion of DNA probes of 40 bacterial microorganisms collected from the Zr compared to Ti abutment groups. It also compares the peri-implant soft tissues and vertical bone changes between the two groups. Significant differences between the two groups would indicate future selection of the abutment and crown types, which have better biologic and esthetic outcomes in the esthetic zone.

2. At present, no index has been devised to assess the subjective and objective esthetic parameters of definitive crown and the surrounding soft tissues. The innovation of a simple scale that incorporates the percentile of the PES/WES score (objective esthetic parameter) and the patients' evaluations of the esthetic outcome (subjective esthetic parameter) of a final crown will provide the clinician with an assessment tool for the treatment esthetic outcomes and improve patients' awareness regarding the standards of esthetics. It is proposed that this scale will be termed the subjective and objective esthetic classification (SOE).

3. This study will be among the first to assess a comprehensive set of variables of clinical relevance in a clinical setting, under investigation, and in a selected cohort of patients.

Research Design and Methods

This project was a continuation of a prospective randomized clinical trial started by the Division of Regenerative and Implant Sciences at the Harvard Dental Center. The clinical trial compared Zr abutments/porcelain fused to Zr crowns and the prefabricated Ti abutments/porcelain fused metal crowns on single implants in the esthetics zone according to biological parameters, esthetic outcomes, and complication rates. Outcome parameters included bacterial colonization, peri-implant soft tissue parameters, vertical bone changes, esthetic outcomes, and patients' satisfaction with the esthetic outcomes, as well as mechanical, biological, and technical complications identified in the two treatment groups throughout the duration of the study.

Study design

Ethical approval was gained from the institutional review board of the Harvard Medical School/Harvard School of Dental Medicine Committee on Human Studies. Patients were recruited from Harvard Dental Center. A 12-month period was allowed for patient enrolment by clinicians in the Oral Implantology Program. Subjects that would benefit from an implant-supported single-tooth restoration in the esthetic zone were included if they met the following criteria:

Inclusion Criteria

General inclusion criteria were patient age ≥ 21 years, the opposing dentition were natural teeth or fixed restorations on teeth or implants, absence of relevant medical conditions and periodontal diseases. In addition, there were local inclusion criteria that included one missing tooth in the esthetic zone, presence of two intact adjacent teeth that were either non-restored or had minor restorations, sufficient amount of bone to achieve primary stability, adequate band of keratinized mucosa (at least 2mm), and adequate oral hygiene. An esthetic zone was defined as any area that was visible in the patient's full smile [57].

Exclusion criteria

General exclusion criteria were presence of conditions requiring chronic routine prophylactic use of antibiotics (e.g., bacterial endocarditis, cardiac valvular anomalies, history of rheumatic heart disease, prosthetic joint replacements), medical conditions requiring prolonged use of steroids, history of radiotherapy or chemotherapy to the head or neck area, physical disabilities that would conflict with the capability to perform adequate oral hygiene, heavy smoking (> 10 cigarettes/day) and inadequate oral hygiene. Furthermore, specific exclusion criteria were missing adjacent tooth, presence of an adjacent implant, presence of periapical pathology at the adjacent teeth, and presence of local inflammation, including untreated periodontitis, persistent intraoral infections, and untreated mucosal diseases.

An informed consent confirmed by the Committee on Human Studies -Harvard School of Dental Medicine was obtained for all the subjects enrolled in this study. Thirty subjects were randomly assigned equally to the two treatment options at the end of the screening visit. A random permuted blocks approach was used to allocate patients into one of the treatment groups. A sealed envelope containing the treatment characteristics (control or test group) was assigned to each patient. Subsequently, and according to the assigned treatment. Zr abutments (Etkon abutment, Straumann AG. Basel. Switzerland)/porcelain fused to Zr crowns or prefabricated anatomic Ti abutments (Straumann AG, Basel, Switzerland)/porcelain fused metal crowns were fabricated and delivered to a clinician (Figure 1). Oral hygiene instructions were given to the patients to improve their hygiene performance. Alginate impressions (DENTSPLY Caulk, Milford, DE) were taken to fabricate diagnostic models for treatment planning.

Treatment procedures and parameters evaluation

Two medical teams were involved in this study: (1) the treatment team, who provided dental care to patients and (2) the investigational team, who evaluated and collected the study parameters/data during the follow-up visits:

Treatment team

After a prosthetic-driven treatment plan had been completed, the treatment team a bone level implant was placed under local anesthesia in each patient according to the surgical protocol (Straumann AG, Basel, Switzerland). After the implant placement standardized periapical radiographs were taken with customized x-ray holder devices. The implant fixture was considered successful if it met the success criteria of Buser et al. [58]. Baseline measurements were by six weeks after implant placement defined as the measurements taken during the healing period after insertion of bone level implant. At the same visit, fixture level impressions (Aquasil Ultra, DENTSPLY Caulk, Milford, DE) were taken to fabricate a screw-retained provisional resin crown. During this phase, patient used removable interim prosthesis until the fixed interim crown was fabricated. The interim crown was delivered three months after the implant placement. One month later, final fixture level impressions were taken to fabricate the final prosthesis according to the assigned treatment groups. The CAD/CAM system was used to fabricate customized Zr abutments with chamfer preparations to have a butt joint with the future crowns. The crown margins were positioned 1-1.5 mm sub-mucosally at the visible buccal areas and less than 1 mm submucosally lingually to improve esthetics and facilitate cement removal. The dimensions of the prefabricated anatomic Ti abutments were selected after trial of different options using the prosthetic planning kit (Straumann AG, Basel, Switzerland). The margins of each crown were positioned 1-2 mm sub-

mucosally. One dental laboratory was used to fabricate all the restorations. At the crown insertion visit, the abutment was torqued up to 35 Ncm according to the manufacturer guidelines, and the crown was cemented using Tempbond[®] temporary cement (Kerr, Orange, CA). The measurements for the crown insertion were taken within a week after the crown delivery.

Investigational team

For subjective evaluations, the trial was designed on the single-blinded level, as the clinicians who evaluated the patients after the crown insertion did not know which the treatment group the patient was in. In addition to the crown insertion visit, three time points were selected to evaluate the study parameters. Time points were one-month, six-months and one-year after crown delivery. At the six-month follow-up, the author joined the investigational team to collect data on the six-month and one-year follow-up visits and to lead the data analysis and interpretation of results. The author attended training sessions to review the objectives of the study and its protocol and to standardize the methods of measuring the study outcomes. Study parameters evaluated during the study included biologic parameters, esthetic parameters, patient's centered-outcomes, and complications.

1. Biologic parameters

Biologic parameters included the evaluation of adjacent teeth plaque score (TPS), implant-prosthetic complex plaque score (IPT), adjacent teeth bleeding score (TBS), and implant-prosthetic complex bleeding score at each visit. Also, plaque samples were collected to examine the microbial colonization around the tested groups and the adjacent teeth. Additional biological parameters were evaluated starting from the baseline visit, including the width of facial keratinized mucosa and probing depth around tested groups and adjacent teeth. Also, the vertical bone changes around the implant-prosthetic complex in the tested groups were measured using standardized periapical radiographs.

The plaque and bleeding scores were reported according to their presence or absence buccally and lingually on the mesial, middle, and distal aspects of the adjacent teeth and the implant-prosthetic complex. The width of keratinized facial mucosa was measured using a periodontal probe (Hu-Friedy, PCP, Chicago, IL) from the zenith of the mid-facial soft tissue at the final crowns and adjacent teeth to the mucogingival junction. Probing depth was done buccally and lingually on the mesial, middle, and distal aspects of the adjacent teeth and the implant-prosthetic site using a periodontal probe. However, the biologic parameters at the implant site were evaluated around the healing screw during the baseline visit.

Standardized periapical long cone parallel radiographs of the implant were taken according to the European Association for Osseointegration Guidelines

[59] to assess the vertical bone changes. To standardize the radiographs between the visits, a film holder was attached to a custom-made bite splint for each patient. From these radiographs, mesial and distal vertical bone changes were measured as a distance from the implant abutment junction to first bone-to-implant contact (DIB) using ImageJ[®] software (National Institutes of Health, Bethesda, MD). Also, the mesial and distal marginal bone heights were measured as distance from the implant abutment junction to the most coronal height of the proximal bone of the adjacent teeth (Figure 2). The implant abutment junction was used as a reference point to obtain all the radiographic measurements. To overcome any magnification problem, the image scale was adjusted by using the implant length, according to the manufacturer's protocol, as a reference.

Plaque samples were collected from the deepest pocket of an adjacent tooth next to the edentulous space at the screening visit. The second samples were collected from the adjacent tooth and the healing screw at baseline before prosthetic rehabilitation. Additional samples were taken at crown insertion visit, one-month, six-months, and one-year after the insertion of the final prostheses. Sample tubes preparation and the microbiological analysis were done in The Forsyth Institute (Cambridge, MA):

Microbiology assessments

Tubes preparation:

Sterile Eppendorf tubes (2 ml) were filled with 100 µl TE buffer (Illumina, Madison, Wl) and placed in a box. The TE buffer was composed of 10 mM Trisbase (pH 8.0) mixed with 0.1 mM ethylenediaminetetraacetic acid (EDTA). It was used to solubilize the DNA and prevent its degradation. Before each visit, two tubes were taken from the samples box and labeled as follows: "T" indicated the plaque sample collected from the deepest pocket of a tooth adjacent to the implant, while "I" indicated the plaque samples obtained from the deepest pocket around the implant or the implant-prosthetic complex. Additional codes were used to specify the visits as follows: "A" for the screening visit, "B" for the Baseline visit, "C" for the crown insertion visit, "D" for the one-month follow-up visit.

Sampling procedure:

After measuring the probing depth around the implant and the adjacent teeth, the deepest probing depth was the site used to obtain the plaque sample. When more than one site had the same depth, the mesial site was used. Each plaque sample was immediately placed in the TE buffer within the Eppendorf tube, and the tube was directly placed in a cooler with ice then kept in a freezer at -80°C until it was transferred to the Microbiology Department at The Forsyth Institute.

Sample Analysis:

The samples were analyzed to detect 40 commensal and pathogenic bacteria frequently detected in gingival samples (Table 1). Analysis of the plaque samples took place at The Forsyth Institute (Cambridge, MA) according to the following protocols composed of four main steps: bacterial cell lysis, DNA clean up, DNA amplification, and checkerboard DNA-DNA hybridization.

DNA was purified from plaque sample using a MasterPure DNA Purification Kit (Illumina, Madison, WI). Cell lysis started by thawing the samples completely, and then Ready-lyse Lysozyme (1uL) was added to each sample. Samples were incubated at 37°C overnight. On the next day, the heating block (65°C) was turned on. The 2X T&C Lysis Solution (100ul) was added to each sample, followed by Proteinase K (1ul), and then samples were mixed by vortexing. Samples were incubated at 65°C for 30 minutes and then placed on ice for 7 minutes.

To start the DNA clean-up process, new tubes were prepared with 400ul isopropanol and placed on ice. Also, ethanol (75%) was placed on ice. MPC Protein Precipitation Reagent (120ul) was added to samples and mixed vigorously for 10 seconds using a vortex machine. The debris was centrifuged for 10 minutes at 10,000xg to form a pellet. Samples were placed on ice immediately following centrifugation. Supernatant solutions were transferred to tubes with isopropanol, and the pellets discarded. The tubes were inverted 30-40 times and then placed on ice for 10 minutes. After that, the tubes were centrifuged at 4°C

for 10 minutes at 10,000xg to form DNA pellets. The isopropanol was removed, and the DNA pellet was saved for each sample. The pellet was then twice rinsed with 500ul 75% ethanol. Finally, the DNA was resuspended in 50ul of TE buffer after the pellet had dried.

Because the DNA amount obtained from each sample was small and insufficient for DNA probe analysis the sample DNA was amplified using Ready-To-Go GenomiPhi V3 DNA Amplification Kit (GE Healthcare Bio-Sciences, Piscataway, NJ). The 2x denaturation buffer (10 µl) was mixed with 1 µl of 10 ng DNA. Then 9 µl PCR-grade water was added. Samples were heated to 95°C for 3 minutes then cooled to 4°C on ice to denature the DNA. Then, the Ready-To-Go GenomiPhi V3 cake was reconstituted with the denatured DNA (20 µl). Each amplification reaction was kept on ice prior to incubation at 30°C. The samples were incubated at 30°C for 1.5 hours to amplify the DNA. After that, samples were heated to 65°C for 10 minutes then cooled to 4°C. Heating the samples was required to prevent the degradation of the amplified DNA by inactivating the exonuclease activity of the Phi29 DNA polymerase. The amplified DNA was stored at -20°C until the checkerboard DNA-DNA analysis.

Checkerboard DNA-DNA hybridization was used to detect the presence of bacterial species in patient samples and to calculate their relative proportions in the samples. The nucleic acids from patient plaque samples were attached to a solid support membrane and subsequently hybridized with labeled DNA probes to identify bacteria of interest. The amplified DNA was affixed onto a positively

charged membrane (nylon filter) using a Mini-Slot apparatus. Digoxigenin-labeled probes were applied using the Mini-Blotter 45 and subsequent overnight incubation allowed hybridization of complimentary DNA strands. This method allowed samples to be analyzed for 40 bacterial species and detection was performed by chemifluorescence. The checkerboard DNA-DNA hybridization procedure took four days and listed as follows:

a. Laying Samples onto the Positively Charged Membrane:

Four membranes were used and labeled corresponding to the checkerboard data sheet. The Mini-Slot (Immunetic Inc., Boston, MA) was assembled. A membrane with the label-side face down was placed on the Mini-Slot (lane 1 of the Mini-Slot was lined up with the top of the membrane). Fifteen sheets of Whatman filter paper were cut to15 X 15 cm dimensions. The bottom of the apparatus was attached and screwed together tightly. The 1 ng standard was made by aliquoting 100 μ L of a 10 ng standard into a tube and diluting with 900 μ L of TE buffer.1500 ng were taken from the amplified DNA samples and placed in 1 ml of TE buffer. Then, the DNA samples and the standards were placed in a boiling water bath for 10 minutes. After boiling, the samples were placed on ice while 1 and 10 ng standards were vortexed and directly each standard was placed into lanes 29 and 30 of the Mini-Slot, respectively. The board was moved side to side to allow complete coverage in the individual slots. 10 ng of genomic DNA was equivalent to 10^6 cells. The standards consisted of the 40 different

DNA probe species. Samples were vortexed then each sample was laid into the 28 lanes of the Mini-Slot using transfer pipettes and left for 5-10 minutes for complete absorption. The Mini-Slot was disassembled, and the four corners of sample area were marked to later help with the lining up. The membrane, along with a sheet of Whatman paper, was placed in an UV Stratalinker (1200 joules), and the DNA side faced up to bind the DNA to the membrane. After that, the filter was discarded and the membrane was left on a metal rack for 10-20 minutes in the oven at 80°C. The membrane was then placed in a hybridization bag with an appropriate label and left in a drawer for the next day. All these steps were repeated with the three remaining membranes.

b. Pre-hybridization and Hybridization:

The prehybridization blocked nonspecific areas on the membrane while the hybridization allowed the annealing of complimentary DNA strands. The prehybridization and hybridization solutions were prepared. The membrane was placed in a box and wetted with 2X saline-sodium citrate (2XSSC). Then, the membrane was placed in a hybridization bag, and 35 mL of pre-hybridization solution was added. Bubbles were removed to ensure complete exposure of the membrane to the pre-hybridization solution, and the bag was sealed using the pouch sealer. The membrane was incubated in a 42°C oven for 2 hours. Bubbles were removed again.

Bacterial probes were removed from the -20°C freezer, diluted with hybridization solution and mixed vigorously using a vortex machine. The concentration of each probe was adjusted to make sure that the final volume equaled 150 µL per checkerboard. The probes were boiled for 10 minutes and cooled on ice for at least 5 minutes to prevent the re-annealing of the DNA single strands. Meanwhile, the membrane was taken from the oven, and the Mini-Blotter 45 was arranged. The membrane was placed face down on the Mini-Blotter with the label at lane #45. A piece of plastic wrap, then a cushion were placed over the membrane. The lower part of the board was assembled and screwed tightly. A 145 µL of each probe was taken and the appropriate lane of the boards was filled. Lanes 1, 12, 34 and 45 were skipped and used as reference points. These lanes were filled with hybridization solution. Lane 23 had human DNA to differentiate it from bacterial DNA. The whole board was wrapped with plastic wrap and placed into a plastic bag. Distilled water (50 mL) was added into the bag to avoid drying out the membrane. The board was left in the oven at 42°C overnight.

c. Washing:

The purpose of the washing was to remove the excess unhybridized probes from the membrane. A 9 L phosphate (PO_4) buffer was added into the circulating water bath. A circulating water bath was turned on and left until the temperature reached 68°C. Meanwhile, 3 L of PO_4 buffer was kept on a heating

plate for a solution change later. The checkerboard was removed from the oven and the excess unhybridized probes were aspirated with an activated vacuum source and the checkerboard was disassembled. The membrane was placed in the circulating water bath at 68°C for 20 minutes. After that, 1/3 of the solution was poured off in the circulating water bath and replaced with the 3 L of the heated PO₄ buffer. After the solution reached 68°C, incubation was continued for another 20 minutes.

During the second PO₄ buffer wash, 9 mL of Tween 20 (Sigma-Aldrich, Saint Louis, MO) was added to maleic acid buffer and blocking buffer was made. 50 mL per membrane was made for use it in the antibody solution and was kept in a separate small flask. Buffer 3 was prepared by mixing diethanolamine solution (50 mL) and magnesium chloride solution (50 mL). The membrane was removed from the circulating water bath, and washed once with maleic acid buffer for 1 minute. Then, the membrane was incubated with the blocking buffer (300 mL per box) for 1 hour on rotator at room temperature. A 3.3 µL (per membrane) of Anti-Dig conjugated alkaline phosphatase 1:15,000 (Roche Life Science, Indianapolis, IN) was added to the small flask of blocking buffer to make the antibody solution. They were mixed for at least 5 minutes to allow complete integration of the antibody. After an hour in the blocking buffer, the membrane was placed into a hybridization bag, and 50 mL of the antibody solution was added on the DNA side of the membrane. Bubbles were removed; the bag was sealed and left on a rotator for 30 minutes at room temperature. Then, the

membrane was washed quickly with the maleic acid buffer (1 minute) to remove the antibody solution. It was then washed with maleic acid buffer for an additional 15 minutes. The last step was repeated three times.

After that, the membrane was washed with Buffer 3 for 5 minutes. Then, 1 mL of ECF[™] Substrate (Sigma-Aldrich, Saint Louis, MO) was diluted in 4 mL of buffer 3 per membrane. The membrane was placed in a small plastic box, and 5 mL of the diluted ECF[™] Substrate was added to cover the membrane completely and evenly. The membrane was then placed in a plastic reaction folder, and air bubbles were removed by wiping the reaction folder gently with a paper towel. The reaction folder was placed onto aluminum foil lined with plastic wrap. The plastic wrap and the foil were wrapped over the folder so that no light could enter. Then, it was stored overnight at room temperature. All these steps were repeated for the remaining three membranes. The checkerboard process had finished at this point, and the output of the results was displayed on the Typhoon Trio scanner (GE Healthcare Bio-Sciences, Pittsburgh, PA).

d. Using Typhoon Trio scanner:

This process allowed quantification of the signals given on the checkerboards by determining the fluorescence intensity. The membrane was positioned in the upper right corner of the reaction folder, distilled water was added, and bubbles were removed. The Typhoon Trio scanner control v5.0 was opened, and the following setting was used:

Templates → Load: checkerboard1 Acquisition Mode: Fluorescence Tray: User Select Pixel Size: 200 (microns) Focal Plane: Platen Image Analysis: Imagequant Orientation: - Setup: Filter: 520 BP 40 Cy2, ECL + Blue FAM (Emission) PMI: 350 (volts) Laser: Blue Laser (488 nm) (Excitation) Sensitivity: Normal

- Press Sample: leave the box unchecked

Distilled water was added on the scanner. The membrane was placed on the Typhoon Trio scanner within the highlighted checkerboard1 template ($B \rightarrow I$ and $2 \rightarrow 9$ on the scanner grid). The membrane faced down with the label in the upper left corner. The surface was smoothed out, bubbles were removed, and distilled water was moved away from the edges of the Typhoon. The cover of the Typhoon was closed to begin scanning. The appropriate folder and the file name were selected. The file name always had the extension ".gel" at the end. After saving these changes, the Typhoon automatically started scanning. Five minutes later, the scanner completed the scanning process; the image of the membrane (Figure 3) appeared and already saved according to the selected folder. Images were analyzed in Phoretix program (TotalLab Limited, Newcastle, United Kingdom). Briefly, a grid was drawn around the membrane spots, the background was subtracted, and then a comparison of the fluorescence intensity between the standard and the samples was calculated. The outcomes were expressed in bacteria x 10^5 and saved in an Excel sheet.

Bacterial prevalence was calculated as the presence or absence of each bacteria species in each sample. The detection level of the bacterial prevalence was set to be equal or more than 1×10^5 to avoid false positive readings. In addition, the total DNA count for each bacterial species was divided by the total count of the DNA in the sample to calculate the proportion of the bacterial DNA probe.

2. Esthetic parameters

The esthetic outcomes in this study were evaluated from two perspectives: clinician evaluation as an objective parameter and patients' evaluation as a subjective evaluation. Starting from the crown insertion visit, the clinician evaluation was performed using the PES and WES at each time point [44]. As described by Belser et al. [44], the PES represented the esthetics of the soft tissue around dental implant-prosthetic complex and included five esthetic parameters: level of the facial mucosa, curvature of the facial mucosa, mesial papilla, distal papilla, and soft tissue color and texture/root convexity at the implant site from the facial aspect. The mesial and distal papillae were assessed for their absence (score 0), incomplete presence (score 1) or complete presence (score 2). The level of the facial soft tissue was compared to the contralateral tooth. A value of 2 indicated identical vertical level, and 1 represented slight discrepancy (\leq 1mm), while 0 was used for major discrepancy (> 1 mm). The curvature of the facial mucosa was compared to the control natural tooth. If the curvature of both sites were identical a score of 2 was given. Otherwise, a score of 1 was reported if they were slightly different, and a score of 0 was given for markedly different conditions. As the soft tissue color and texture/root convexity combined three different elements, scores were given as follows: 2 when all three elements were identical to control tooth, 1 when two elements were present, and 0 when only one element was fulfilled or when none of them were

fulfilled. A value of 2 represented the best esthetic outcome and clinically acceptable total PES should have a value of 6 or more.

The WES evaluated the esthetic outcome of the final clinical crown. The general form of the tooth; volume and outline of the visible crown; color (value and hue); surface texture; characterization and translucency were the five esthetic variables incorporated into WES. For each variable, a score of 0, 1, or 2 was given according to the degree of discrepancy when compared to control tooth. A value of 2 indicated no discrepancy, 1 indicated minor discrepancy, and 0 indicated major discrepancy. Total WES would be equal to 6 or more if the prosthesis was esthetically acceptable clinically.

The total esthetic score (PES/WES) was the sum of the total PES and the total WES. The highest possible total PES/WES was 20, which indicated a close proximity of the peri-implant soft tissue parameters and the clinical implant crown parameters to the corresponding features present at the contralateral natural tooth.

Changes in mesial and distal papillae heights of the implant crown, crown height, or mucosal recessions of the implant crown and the adjacent teeth were measured to objectively evaluate the differences in soft tissues before and after the implant crown insertion according to Buser et al. [60]. Briefly, impressions were taken at each designated time point of the study to produce casts of type IV stone (Whip Mix, Hamden, CT). These measurements were done to assess the soft tissue around the implant crown as follows:

Papilla height was the distance between the tangent line of the zenith of the mid-facial gingival margin of the adjacent teeth to the most coronal part of the mesial or distal papilla (Figure 4).

Clinical crown height was the distance between the zenith of the mid-facial gingival/mucosal margin and the most coronal part of incisal edge or occlusal surface of adjacent teeth and the implant crown (Figure 4). Comparisons between the crown height measurements at each time point were used to assess the facial gingival/mucosal recession or overgrowth. The implant clinical crown height was recorded as "0" at the baseline [60]. All the esthetic parameters were recoded by 2 clinicians and the accuracy of measurements was confirmed by another clinician in the investigational team during all time points except for the one year follow-up where the measurements were recoded and confirmed by the same clinician.

3. Patients' centered-outcomes

Patients were asked to fill out a questionnaire to assess their self-esteem and the esthetic outcome of the treatment provided in both abutment groups tested to record their subjective evaluations. All the questions were presented in VAS format. Patients were asked to draw a vertical line on a calibrated horizontal line (100 mm) to score their answers [44, 51]. At the baseline, patients were asked to rate the impact of tooth loss on their self-esteem and their expectations regarding the final esthetic outcome. The scale of the impact of tooth loss ranged from "not affected at all" to "very much affected", while the scale associated with the patient expectation regarding the final esthetic outcome ranged from "low" to "high".

At the crown insertion visit, patients were asked to rate the esthetic accomplished by the treatment and rate their satisfaction. The scale of the esthetics accomplished ranged from "not at all fulfilled" to "completely fulfilled" while the scale associated with the patient overall treatment satisfaction ranged from "not at all satisfactory" to "completely satisfactory".

At each follow-up visit, patients rated their overall treatment satisfaction, impact of the treatment on their self-esteem, and the esthetics of the restoration provided. The same scales were used to record the patients' answers in the follow-up visits to allow for the comparison of the results before and after the treatment.

The total PES/WES score and the level of a patients' esthetic evaluation were used to develop an experimental percentile scale called the subjective and objective esthetic classification (SOE) for definitive crown evaluation. Based on the obtained percentages, the final prosthesis and the adjacent soft tissues were classified into the following categories to assess the success of the treatment esthetic outcome:

- Satisfactory: when patient's esthetic evaluation was ≥ 80% and the sum of PES/WES was ≥ 60%. Prosthesis in this category can be delivered as it meets the required esthetic outcome, as well as the adjacent soft tissues from both the clinician's and the patient's perspective.
- Marginal: when patient's esthetic evaluation was ≥ 50% but < 80%, and/or the sum of PES/WES was ≥ 50% but < 60%. Prosthesis that falls in this category needs further clinical and/or laboratory modifications before delivery. The clinician should explain to the patient the reasons for further modifications of the prosthesis to obtain a satisfactory esthetic outcome.
- Unsatisfactory: when both patient's esthetic evaluation and PES/WES score were < 50%. Prosthesis that falls in this category needs reconstruction of the prosthesis and/or complex soft tissue management.

The VAS scores usually range from 0 -100. A score of 50 usually represent the average value obtained. Therefore, a threshold of 50 was used to set the difference between the unsatisfactory category and the marginal category

for patients' esthetic evaluations. A threshold of 80 was used to set the difference between the marginal category and the satisfactory category for patients' esthetic evaluations to accommodate for their high esthetic expectations. Similarly, modifications of the PES/WES index were set for the threshold of the SOE categories from the clinicians' evaluation. First, the value of the PES/WES was changed to a percentage. Second, a new threshold point was added at 50%, the average value, to set the difference between the unsatisfactory category and the marginal category. Finally, the percentage of the clinical acceptance threshold proposed by Belser et al. was 60% [44], which was used to set the difference between the marginal category and the satisfactory category.

4. Complications

All the implants were examined for the success criteria of Buser et al., including absence of a continuous radiolucency around the implant, mobility, recurrent peri-implant infection, and persistent subjective complaints, such as foreign body sensation, pain, and/or dysesthesia [58].

The incidence of any biological, mechanical, and technical complications for the implant-prosthetic complex was reported. Biological complications included issues that involve the soft tissues, such as fistulas, suppuration, bleeding, gingival inflammation, and soft tissue dehiscence. Mechanical complications were defined as issues that involve the implant-prosthetic complex, such as debonding of the crown from the abutment, abutment screw loosening, abutment screw, and/or fracture of the implant. Technical complications were related to laboratory procedures and fabrication techniques, such as fracture of the veneer material, fracture of the crown fit. In a case of any complication, treatment was provided according the current standards of care. The prevalence of these complications was reported and compared between the two abutment groups.

Statistical Analysis

Statistical analyses were performed using SAS 9.4 software (SAS Institute, Cary, NC). Histograms were created for the tested variables, and illustrated that the data was not normally distributed. A significance level of α = 0.05 was used, except for the bacterial analyses, for which a significance level of P < 0.00128 was computed to compensate for the multiple comparisons for the 40 bacterial species as described by Socransky et al. [61]:

Overall P value of $0.05 = 1 - (1 - k)^{40}$ where k was the desired individual p-value.

Descriptive statistics were calculated for TPS, IPS, TBS, IBS, width of facial keratinized mucosa, probing depth, bacterial species, and vertical bone changes around the implant-prosthetic complex and the adjacent teeth. A Chi-square test was used to test for equal detection frequency of the bacterial species (i.e. prevalence of the bacterial species) between the two abutment groups. When the expected cell frequency was less than 5, the p-value of the Fisher's exact test was used instead of the Chi-square. The Mann-Whitney U test was used to evaluate differences in the bacterial proportions around the implant-prosthetic complex and the adjacent teeth between both groups. Comparison of the TPS, IPS, TBS, IBS, width of facial keratinized mucosa, probing depth, and vertical bone changes between the two groups was done longitudinally at the different time points by using linear mixed effects models for continuous outcomes as described by Gallucci et al. [18].

Descriptive statistics were computed for PES parameters, WES parameters, total PES/WES, papillae heights, buccal gingival margin, buccal peri-implant mucosa margin and all VAS scores. All these variables were compared longitudinally by using the linear mixed effects models for continuous outcomes. The SOE scale between the percentage of the total PES/WES scores and the percentage of patients' scores from VAS of overall treatment satisfaction was established. Based on the obtained percentages, the final prostheses and the adjacent soft tissues were classified into three categories: satisfactory, marginal and unsatisfactory. Then, a Fishers exact test was done to test for equal proportions of SOE outcomes between the two tested groups. Implant-prostheses complication prevalence was reported according to the incidence of various complications.

Results

Subject population and follow-ups

30 patients were involved in this study and evenly allocated to the Ti and Zr groups. One patient from the Zr group decided to withdraw after the screening visit. Thus, the participants in the study consisted of 15 (51.72%) females and 14 (48.28%) males. The mean age at the screening visit was 45.03 ± 13.77 years and the range was 22 - 73 years. The implant sites included 11 central incisors, 9 lateral incisors, 1 canine, and 8 first premolars. According to the prosthetic-driven treatment plans, the implant length selected was 8mm in five cases (17.24%), 10 mm in 17 cases (58.62%) and 12 mm in seven cases (24.14%). Implant diameters were 3.3 mm in 13 cases (44.83%) and 4.1 mm in 16 cases (55.17%).

During the study, some patients failed to show-up for all follow-up visits. At the one-month follow-up visit, one patient from each group failed to show-up. At the six-month follow-up, one patient from each group did not show-up. However, three patients; one from the Ti group and two from the Zr group, did not show up for the six-month and one-year follow-ups as one moved to another state and two became unresponsive to phone calls and mail notices. Finally, only one patient from the Ti group did not show-up at the one-year follow-up visit.

Plaque and health of soft tissues

Generally, patients in both groups maintained adequate oral hygiene. The plaque score ranged from 1-5 around the adjacent teeth and 1-4 around the implant-prosthetic complex. Moreover, the bleeding score ranged from 0-3 around the adjacent teeth and 0-2 around the implant-prosthetic complex. The width of facial keratinized tissues was 2-10 mm and 2-8 mm around the adjacent teeth and the implant-prosthetic complex, respectively. The probing depth was 1-4 mm around the adjacent teeth.

The adjacent teeth plaque and bleeding scores, implant-prosthetic complex plaque bleeding and score, width of keratinized tissue and probing depth measurements did not show any statistically significant differences when the mean values were compared at each time point between both groups (Table 2). The only exception was the mean probing depth of the mesial tooth at the mesio-lingual site (p= 0.02), which was less in the Zr group.

Radiographic vertical bone changes

At the baseline, the radiographic images showed the first bone to implant contact on both sites were approximately at the implant shoulder level. However, the position of this bone slightly moved apically during the time points and by the one-year follow-up it returned slightly coronal position (Figure 5). All the proximal margin bones were located above the implant shoulder at the baseline visit and this level was maintained through all the study period except for the distal

marginal bone in the zirconia group, which was decreased slightly (Figure 6). However, no statistically significant differences were detected when the mean values of the vertical bone changes were compared at each time point between both groups.

Samples for microbial analysis

Within the Ti group, eight samples failed to yield sufficient DNA for analysis. Two samples were from the screening visits and six from the one-year follow-up visit. The lost samples from the initial visit were substituted by a sample obtained from the adjacent tooth at the baseline visit before the implant placement and a sample taken from the adjacent tooth before the placement of the final prosthesis in the crown insertion visit. The lost samples from the final visit were substituted by samples taken from six-months follow-up visits. For the patient who did not show-up at the one-year follow up visit, the six-months sample was used. Two samples from the one-month follow up visits.

In the Zr group, five samples failed to provide good DNA and comprised two samples from screening visits and three samples from the one-year follow-up visits. The lost samples from the initial visit were substituted by a sample obtained from the adjacent tooth at the baseline visit before the implant placement and a sample taken from the adjacent tooth before the placement of the final prosthesis in the crown insertion visit. The lost samples from the final

visit were substituted by samples taken from six-months follow-up visits. For those who did attend the six-month and one-year follow-up visits only one sample from the one-month follow-up was analyzed while the other failed to yield sufficient DNA.

Microbiology results

No statistically significant differences were detected for the mean prevalence of the bacteria between the teeth plaque samples in both groups at the initial and final visits (Figures 7 and 8). However, the mean prevalence of the bacteria around the Zr abutment was significantly greater than the Ti abutment at the final visits for *Streptococcus intermedius* (p< 0.0001), while the mean prevalence of remaining bacterial species was not different (Figure 9).

The mean proportions of the DNA probes of the teeth and the abutment plaque samples in both groups at the initial and final visits showed no statistically significant differences. However, only the mean proportion of the *Treponema denticola* DNA probe (p= 0.0007) was significantly less in the Zr group than in the Ti group at the final visit (Figure 10).

Objective and subjective esthetic outcomes

The means of PES, WES, and total PESWES reflected clinically acceptable outcomes for both treatment groups. Between the group analyses for

the PES parameters, WES parameters, total PESWES score, papillae height, clinical crown length of the adjacent teeth, and the implant crown, there were no statistically significant differences observed when the mean values were compared at each time point (Figures 11, 12, and Table 3).

In general, tooth loss in the esthetic zone affected the patients' selfesteem in both groups without a statistically significant difference (p= 0.66) when the mean values were compared at each time point. In the baseline visit, the answers of four patients only in the Ti group were less than 80% with a range of 2-100% and a mean of 65.25 ± 40.91 . However, the answers of seven patients in the Zr group were less than 80% with a range of 0-100% with a mean of $65.82 \pm$ 37.05. By the end of the treatment at the one-year follow-up visit, the mean scores for the impact of the treatment on the patients' self-esteem were $86.23 \pm$ 27.33 and 96.17 ± 6.24 in the Ti and Zr groups respectively. Only three patients rated the impact on the treatment on their self-esteem less than 80% in the Ti group (Figure 13).

All the patients involved in this study had high expectations for the treatment esthetic results, as they scored mean values of 93.08 \pm 10.49 and 89.35 \pm 15.43 for the Ti and the Zr groups, respectively, at the baseline visits. The final esthetic results accomplished in both groups rated by the patients had a mean value of 95.08 \pm 8.02 and 93.33 \pm 11.13 in the Ti and Zr groups, respectively. There was no statistically significant difference between both groups when the mean values were compared at each time point (Figure 14).

There was no statistically significant difference in the overall treatment satisfaction between both groups when the mean values were compared at each time point. Overall, the means of the overall treatment satisfaction in Ti and Zr groups were higher than 80% of the VAS through all the visits (Figure 15).

According to the SOE classification, the final prostheses and the adjacent soft tissues in both groups fell under the satisfactory category except in five incidences where they were categorized as marginal (Figure 16). The Ti group was classified as marginal in three occurrences because the VAS esthetic score was rated below 80% in three visits. On the other hand, the Zr group was in the marginal category in two incidents reflecting that the VAS esthetic score was below 80% in two visits. In the satisfactory category, the mean PES/WES was 81.05 \pm 11.32, while the mean score of VAS regarding the esthetic of the treatment was 96.85 \pm 4.59. In the marginal category, the mean PES/WES was 78 \pm 10.37 while the mean score of VAS regarding the esthetic of the treatment was 67.40 \pm 10.69. However, there was no statistically significant difference found between the both groups in the distribution of the SOE categories (p= 0.67).

Complications

Although some complications were reported, the success rate for the implant fixtures was 100% in both groups. Only two patients in the Ti group had biological complications at the crown insertion visit that involved mucosal

inflammation (1.19%) and soft tissue dehiscence at the mesial papilla (1.19%). Four patients in the Zr group complained once of mucosal inflammation (5.06%) at the crown insertion visit, one-month, six-month and one-year follow-up visits.

Mechanical complications involved debonding of the crown form the abutment and abutment screw fracture. Debonding of the crown occurred twice in two different patients in the Ti group (2.38%) by the time of the one-month and one-year follow-up visits. Abutment screw fracture happened during the delivery of a Zr abutment (1.27%).

Technical complications reported in this study were crown shade mismatch (1.19%) and veneering porcelain chipping (2.46%). Shade mismatch was noticed in the Ti group during initial delivery of the crown, and it was corrected before the final crown delivery. Minor veneer porcelain chipping occurred in one patient in each group at the six-month follow-up visit. One patient in the Ti group required just finishing and polishing to the crown to eliminate a sharp edge, while one patient in the Zr group received a small composite restoration.

Discussion

This study provided a comprehensive comparison between the Ti and Zr abutment groups using various biologic and esthetic parameters. No major biologic and esthetic differences were detected between the abutments groups in the current study. Similar findings have been reported in different studies when all-ceramic abutments and restorations were compared to metallic abutments and porcelain fused to metal restorations [23, 24, 48-50]. Therefore, the hypothesis of the study was rejected for all the variables except for the bacterial prevalence and proportions of selected species, and soft tissue probing depth of the mesial tooth at the mesio-lingual site. This study also indicated that the newly proposed subjective and objective esthetic (SOE) classification was reproducible and would be valuable as an assessment tool for treatment esthetic outcomes.

The plaque scores were low, reflecting good oral hygiene performed by the patients with re-enforcement of oral hygiene instructions at each visit. In the Ti group, the plaque score on implant-prosthetic complex was lower than on adjacent teeth, which is consistent with the findings of Sailer et al. [24]. However, A recent study by Zembic et al. reported slightly higher plaque accumulation around Ti abutments than teeth, but the increased plaque observed was not statistically significant [23]. The Zr group showed a comparable amount of plaque present on implant-prosthetic complex and adjacent teeth after crown placement. This finding is in accordance with Glauser at al. for subjects after one-year of follow-ups but not with the findings of Ekfeldt et al. who reported lower plaque

accumulation on Zr abutments than on natural teeth [16, 25]. Between both groups, Zr group had slightly higher plaque accumulations than the Ti group as was observed by Sailer et al. [24] however the differences were not statistically significant. Differences in plaque accumulation might be due to differences in the emergence profile of both abutments as proposed by Sailer et al. [24].

In general, bleeding upon probing around both abutment types was slightly lower than the natural teeth, except for the visits of the crown insertion and the six-month follow-up in the zirconia group, where it was slightly higher and equivalent to the natural teeth. This contrasted with reports of Salier et al. and Zembic et al. who reported a higher incidence of bleeding upon probing at the abutment sites than with natural teeth [23, 24]. This difference can occur due to the differences in the amount of plaque accumulated around abutments and natural teeth. However, there was no statistically significant difference between groups, which was consistent with the results of Zembic et al. and Hosseini et al. [23, 48, 49].

The width of buccal keratinized mucosa was similar between the implantprosthetic site and the adjacent teeth in both groups. Gallucci et al. reported a similar width of keratinized mucosa when alumina abutments were compared to metallic abutments at 2-years of follow-up [50]. Positioning the crestal incision lingually and reflecting the keratinized mucosa buccally during the time of implant placement led to optimizing width of keratinized mucosa for implants [50]. The

integrity of the keratinized mucosa was not affected by the use of different types of abutments in this study.

The probing depth at the adjacent teeth in the Ti group ranged from 1-3 mm, while in the Zr group was within 1-4 mm. Although there was a statistically significant difference between both groups at the mesio-lingual site at the mesial tooth, it was not clinically significant as the probing depth was within the normal healthy range. This finding is consistent with Gallucci et al., where the probing depth of the adjacent teeth did not exceed 5 mm when ceramic abutments were used [18].

The mean of the distance from the implant abutment junction to first boneto-implant contact was approximately at the implant shoulder level at the baseline visit on the mesial and distal sites in both groups. However, bone level at a slightly apical direction was observed at crown insertion, one-month, and sixmonths follow-up visits, but the bone level was slightly coronal at the one-year follow-up visit. However, all the DIB values represented good bone height around the implants in both groups, as bone loss did not exceed the 1.5 mm bone loss reported by Papaspyridakos et al. [47]. The bone remodeling process to establish the peri-implant biological width is probably the reason behind the observed changes in bone height. The current study results were consistent with report of Buser et al. that observed changes in DIB values increased around dental implants in single-tooth gaps located in the maxillary esthetic zone at different time points after crown insertion [60]. Also, there was no statistically significant

difference in the current study between both groups for DIB values, which was similar to findings by Hosseini et al. after comparing the clinical performance of Zr and Ti abutments over one year [48]. The changes in the mesial and distal marginal bone level were not statistically significant between both groups. Minimal bone level changes may occur due to the pressure created on the soft tissue by the crown placement [18]. Zembic et al. reported that there were no significant differences between Zr and Ti abutments in peri-implant marginal bone levels after 5-years of follow-up [23].

In both groups, the 40 bacterial probe species were detected in the amplified samples collected from the adjacent teeth and the implant prosthesis. The prevalence of the bacteria species between the adjacent teeth was not different between initial and final visits indicating stability of the microbiota over time, including with the imposition of placing implants and restorations. The microbiota of abutment groups differed only for increased detection frequency for *Streptococcus intermedius* in the Zr group compared with the Ti group. Since *Streptococcus* species are generally detected in gingival health, this observation would not reflect a pathological change. Further there was no difference in healthy gingival status between groups suggesting that there were no biologically significant differences. It is possible, however, that slight differences might reflect the presence of a slightly higher level of plaque around Zr abutments than Ti. A study by Rimondini et al. reported higher accumulations of another *Streptococcus* species, *Streptococcus mutans* to Zr disks than Ti disks after 24

hours of exposure to the oral cavity although other bacterial species did not show differences [28]. It will be interesting to examine the adherence of different Streptococcus species on Zr with different degrees of polishing and glazing to improve the quality of the material used in implant dentistry. Other clinical studies did not detect any differences when the DNA counts of Porphyromonas gingivalis, Aggregatibacter actinomycetemcomitans, and total bacteria were evaluated between Zr and Ti abutments [35, 36]. Furthermore, increased detection of selected species around Zr compared with Ti abutments for overdenture prostheses were observed in edentulous subjects including for Porphyromonas gingivalis, Prevotella intermedia, Tannerella forsythia, Parvimonas micros, Aggregatibacter actinomycetemcomitans, Fusobacterium nucleatum and Treponema denticola [34]. The bacteria detected were similar to those observed by Lee et al. in a study that compared the microbiota of teeth, the dorsum of the tongue and the implant-prosthetic complex [38].

Comparable proportions of the DNA probe species were found in the samples from teeth adjacent to implants and abutment sites from both groups at initial and final visits. Although the mean proportion of *Treponema denticola* was significantly less in the Zr group than the Ti group at the final visit, both groups had proportional levels by far below levels detected in disease, for example, in peri-implantitis [33]. All these biological parameters indicated that the environment around Zr and Ti groups was compatible with maintenance of

healthy soft and hard tissues after one year of functional loading of implants in the esthetic zone.

Both abutments and associated restorations achieved good esthetic scores at each time point during the study using the objective parameters used by the clinicians. When the objective esthetic parameters were compared no differences were detected between the two groups. These observations were in accordance with a recent study by Carrillo de Albornoz et al. [42]. This might be due to the presence of healthy soft tissues around the abutments, proper width of keratinized tissues, and good soft tissue morphology from having used the appropriate dimensions for the interim and final prostheses [18, 50].

In general, the changes in PES followed the changes in the soft tissues after the insertion of the crowns. The height of the interproximal papillae slightly increased in both groups at the one-year follow-up visit in comparison to the baseline and crown insertion visits. Similar findings were documented by Gallucci et al. when the dimensional changes of peri-implant soft tissue of single-implant crowns in the anterior maxilla were analyzed [18]. Although there was no statistical significant difference between both groups in crown height, the Zr group had slightly longer crown heights than the Ti group. This was reflected by the scores given to the level of facial mucosa in PES and the slightly lower values observed for the Zr group. Findings were consistent with results of Gallucci et al. when ceramic abutments were compared to gold abutments [50]

and may have resulted from differences in the emergence profile of different abutments at the mucosal margin.

Since the same dental laboratory fabricated the implant crowns, the WES was similar and clinically acceptable in both groups. Achieving optimal crown color in the Ti group was the most challenging variable to the technicians making the crowns as color had the lowest scores while the crown surface texture was the easiest to reproduce. In general, there were no differences in WES observed between both groups. An explanation of difference in the WES between visits was proposed by Gallucci et al. [50] namely that implant crown material is a variable among a group of variables required to acquire a balanced esthetic combination. These variables included reflection of light, surface texture, tooth morphology, translucency, simulating the contra-lateral tooth, and proper soft tissue embracement [50].

Although there was no statistical difference detected between both groups in regard to patients' self-esteem, most, but not all patients involved in this study were affected due the loss of tooth in the esthetic zone at the baseline visit. In this small group of patients, the lost teeth were 6 incisors, 1 canine and 3 premolars. This may suggest an absence of a relationship between the position of the missing tooth and the patients' self-esteem. One notable finding was that the self-esteem of most patients increased after crown insertion regardless the type of the prosthesis used. This was also noted by Chang et al. who reported that almost all the patients were confident enough to smile after single-implant

restorations in the anterior maxilla [51]. However, when missing molar teeth were replaced by implants no differences were reported between the immediate and conventional placement protocols [52]. Further comprehensive investigations including different aspects such as age, gender, socioeconomic level, education, occupation and clinical variables are needed to understand the impact of singleimplant therapy on the patients' quality of life.

The missing tooth in this study was located in the esthetic zone and patients' expectations of the final esthetic outcome were high in both groups. However, the scores for the accomplished final esthetic results in both groups met the expectations of patients through the whole study period. Several studies reported high degrees of patients' esthetic assessments for single-implant restorations [44, 48, 50, 51]. Furthermore, the esthetic outcome scored by patients between the restorations in Zr and Ti groups did not differ as observed in other clinical studies [48, 50]. In addition, all patients in the current study showed high satisfaction levels without any noticeable difference between the groups. The reason behind that may be the appropriate biologic and esthetic results achieved by both treatment options.

Another factor to consider is the different standards used by patients to express their esthetic evaluations and overall treatment satisfaction compared with assessments by clinicians [44, 50, 51]. In an attempt to resolve the differences between patients' and clinicians' evaluations regarding a final crown and its surrounding soft tissue, the SOE classification was created. This

straightforward scoring was composed of only three categories to reflect success of treatment esthetic outcomes. These categories were satisfactory, marginal, and unsatisfactory which are easy to memorize. In addition, evaluating a final crown and its surrounding tissues using the SOE depends on two quick simple assessment tools, namely performing PES/WES analysis and evaluating the patient's VAS score to determine the esthetic outcome. These two assessment tools are proven and well documented in the literature [44-46, 48, 50, 51, 60]. The SOE can be performed by most of dentists and require minimal time. Thus, it may help to improve treatment esthetic outcomes, which, in turn, will improve the standards of care provided to patients. However, future prospective studies are required to assess the strengths and weaknesses of this classification and its use as a fundamental part of studies evaluating treatment esthetic outcomes of single implant restorations in the esthetic zone.

In this study, the success of treatment esthetic outcome was within the satisfactory category in both groups except in five incidents where it was marginal. The mean values of the esthetic evaluations performed by patients and clinicians were above 80% in this category, which indicated both evaluating groups agreed that the required esthetic outcome had been achieved. Within the marginal category, although the clinician evaluation indicated acceptable esthetic outcomes, patients' scores were below the satisfactory level. This might be because the expectations of the patients were higher than realistically could be achieved. In these cases the clinician explained to the patients the existing

challenges and the existing solutions. However, there no statistically significant difference was found between both abutment groups in the patients' SOE score categories.

Minor complications occurred through the study period, however, the survival rate for the implant fixtures was 100% in both groups. Mucosal inflammation was found in both groups and treated by prescribing 500 mg of Amoxicillin antibiotic and Chlorhexidine Gluconate (0.12%) mouthwash for 10 days. An additional follow-up visit was made and the inflammation was found have resolved. This observation was as reported by Hosseini et al. who detected an inflammatory response around 7 all-ceramic crowns and 3 porcelain fuse to metal crowns after one year of observation [48]. Another unexpected biologic complication was soft tissue dehiscence at the mesial papilla in the Ti group at the crown insertion visit, assumed to be due to the pressure caused by the crown on the tissue. A crown adjustment was performed and oral hygiene instruction was re-enforced; however, partial loss of the mesial papilla occurred.

Debonding of the crown occurred for Ti abutments that were used to restore a maxillary lateral incisor and a premolar at the one-month and one-year visits, respectively. This resulted from washout of the temporary cement and the small geometry of the abutments used to create porcelain space, which, in turn, reduced the mechanical retention. Both debonded crowns were recemented with the same temporary cement, and no further complications were noticed. A similar result was reported by Hosseini et al., although; in the literature report the crown

and abutment were remade to improve mechanical retention [48]. None of the abutments had complications through the current study. However, one abutment screw was fractured during the delivery in the Zr group and it was replaced with a new one. This was likely due to a problem with the manufacturer and has no relationship with abutment loading. Zembic et al. did not report any Zr abutment or screw fractures after 5 years of functional loading of the posterior implants [23].

A shade mismatch was observed in the Ti group during the initial delivery of the crown, and it was corrected before the final crown delivery. This was observed by Hosseini et al. and reflected that color match is generally easier to achieve when Zr abutments are used [48]. Minor veneer porcelain chipping occurred in both groups during the study. This complication was reported with allceramic implant abutment and crown in Hosseini et al. study with a similar frequency [49].

One major limitation of this clinical study was the small sample size in each group, so study findings may not be reproducible. Other limitations included differences in the soft tissue and bone in the maxillary and mandibular arches, differences in the location of the implant restorations and differences in the sizes of the implants required to restore the missing teeth. Also, different clinicians were involved in the clinical evaluations and plaque sampling throughout the study, which may introduce chances for error. In addition, the one-year follow-up period might be considered as short-term since the long-term integrity of the soft

and hard tissues around the implant-prosthetic complex and esthetic parameters of different groups must be evaluated in longer follow-ups. Therefore, a larger number of randomized controlled clinical studies with large sample sizes and longer follow-up intervals are required to establish evidence in implant dentistry regarding the validity of the relative clinical performance of Ti and Zr abutments.

Conclusion

- 1. Both treatment groups had healthy soft and hard tissues after one year of functional loading. Although statistically significant differences in the probing depth at the mesio-lingual site of the mesial adjacent tooth, and prevalence and proportions of the bacterial colonization were observed, these differences did not affect the integrity of the health of the soft and bone tissues.
- This study revealed no significant differences in the objective esthetic outcomes when CAD/CAM Zr and prefabricated anatomic titanium abutments were used to restore single implants in the esthetic zone.
- Patients' centered outcomes showed highly satisfactory results scored by all patients regardless of the type of the abutment used. In addition, self-esteem of most patients increased after the crown insertion regardless of abutment/implant type.
- 4. The proposed SOE classification is an assessment tool that combined the clinicians' and patients' esthetic evaluations and can be used to assess treatment esthetic outcomes in the esthetic zone.
- 5. In general, Zr group showed comparable results to Ti group after one-year of observation. These results indicated that good biological and esthetic outcomes could be achieved by either treatment option. However, further observations are needed to assess results over a longer term.

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Figure 1: Treatment groups

(A) Ti group, prefabricated anatomic titanium abutment and porcelain fused to metal crown. (B) Zr group, CAD/CAM zirconia abutment and porcelain fused to zirconia crown.



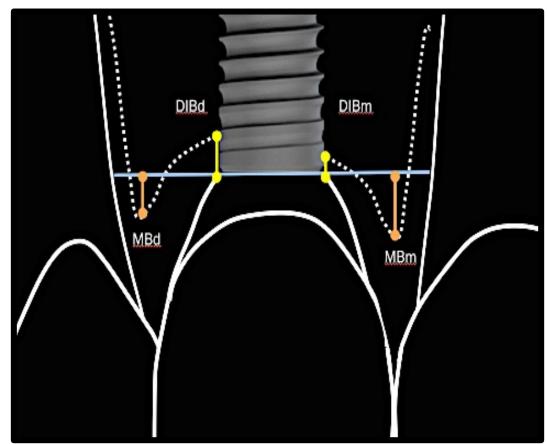


A Ti group

B Zr group

Figure 2: Measurement of the vertical bone changes

The figure illustrates the method used to measure vertical bone heights. The implant abutment junction was used as a reference to measure the distance from the implant shoulder to first bone-to-implant mesially and distally. It was also used to measure the height of marginal bone of mesial and distal adjacent teeth.



DIBm: distance from the implant shoulder to first bone-to-implant mesially.

DIBd: distance from the implant shoulder to first bone-to-implant distally.

MBm: marginal bone of the mesial tooth. MBd: marginal bone of the distal tooth

Figure 3: DNA checkerboard membrane

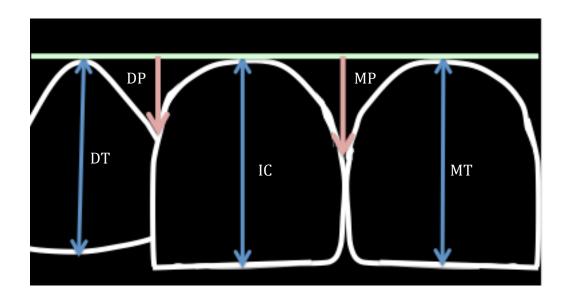
An image from the Typhoon scanner of a DNA checkerboard membrane. The size and intensity of each spot indicates the expression level of the DNA probe in each sample. The last two columns from the right indicate the 10^5 and 10^6 mixed bacterial standards.

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S. anginosus		•		1	٠	٠								1		1		٠	٠	1			٠	1					٠	
S. sanguinis	1	:			4	4		1						4	1	1		1	4	1			1	1			4	1		
A. gerencseriae S. oralis				1	٠	٠	٠				+			٠	٠	•		٠	٠	٠		1		٠	٠					
S. orans C. ochracea		1												5				4	4				4							
A. israelli		1		4			1							5	1	1			۰.	!										
S. intermedius					•						τ.			1	1	•				'	1						5			
T. denticola		1				4												4	4	٠							<u>.</u>			
Human DNA probe		1		1	4	1		4			1				4	1	-	1	1				1				1	1		
P. nigrescens		•		•	•			•	•	•		•	٠		٠		٠	۰	٠	٠		٠		•	٠		•			
A. odontolyticus														÷.																
F. nucleatum ss polymorphum															1			1	1											
C. showae	C						4							х.					4	٠										
F. periodonticum		1			1	1	1	1			1			1		٠		1	х				τ.	4	۰.		÷	- 1		
N. mucosa		1					1							1			•		τ.				1				•			
F. nucleatum ss nucleatum																	٠		έ.				٠	٠						
C. gingivalis		ī	1	÷	1	1	T	- 1			۰.			÷.				1	а.					τ.	Τ.		τ.	- 1		
S. gordonii		1				1	1												÷					1	1		÷			
T. forsythia	C)					1					4								۰.											
		7																	5		0									
S. noxia	6	1		1							1																			1
P. acnes		٠		1												÷		1			1								ī	
P. melaninogenica								1			+					٠		٠	÷	٠			٠					1		
S. mitis		٠			٠		+	- 1			1			٠	1	٠		٠		٠				1			4		÷	
E. corrodens	C				1		1	4			1			8		÷.		٠	ŝ.	٠			ĩ	1				1.1	÷.	
G. morbillorum																														
C. sputigena		1					+				1			4		1		٠	1	1			1							
L. bucallis																														
C. gracilis											3							1							1		1			
P. intermedia			1			1		1										٠	٠					٠	٠		1	1		
																			100											

 $1 \hspace{.1in} 2 \hspace{.1in} 3 \hspace{.1in} 4 \hspace{.1in} 5 \hspace{.1in} 6 \hspace{.1in} 7 \hspace{.1in} 8 \hspace{.1in} 9 \hspace{.1in} 10 \hspace{.1in} 11 \hspace{.1in} 12 \hspace{.1in} 13 \hspace{.1in} 14 \hspace{.1in} 15 \hspace{.1in} 16 \hspace{.1in} 17 \hspace{.1in} 18 \hspace{.1in} 19 \hspace{.1in} 20 \hspace{.1in} 21 \hspace{.1in} 22 \hspace{.1in} 23 \hspace{.1in} 24 \hspace{.1in} 25 \hspace{.1in} 26 \hspace{.1in} 27 \hspace{.1in} 10^5 \hspace{.1in} 10^6$

Figurer 4: Papilla and crown heights

Papilla height was measured as the distance between the tangent line (green color) of the zenith of the mid-facial gingival margin of the adjacent teeth to the most coronal part of the mesial or distal papilla. Clinical crown height was measured as the distance between the zenith of the mid-facial gingival/mucosal margin and the most coronal part of incisal edge or occlusal surface of adjacent teeth and the implant crown.

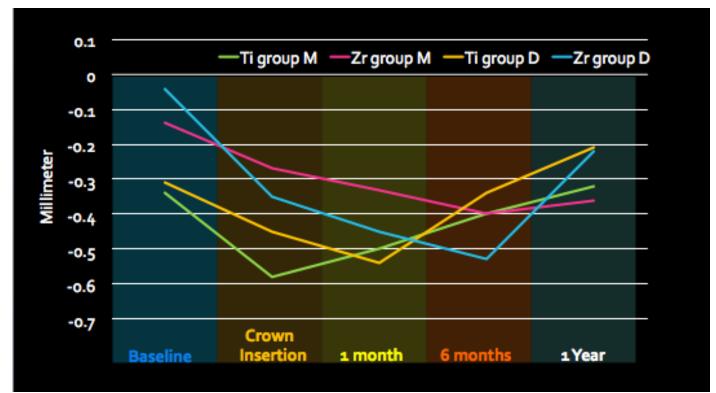


MT: mesial tooth. IC: implant crown. DT: distal tooth.

MP: mesial papilla. DP: distal papilla.

Figure 5: Vertical bone changes (DIB)

Changes in the distance from the implant-abutment junction to bone-to-implant occurred around the implant-prosthetic complex on the mesial and distal sites in both groups throughout the study time points.

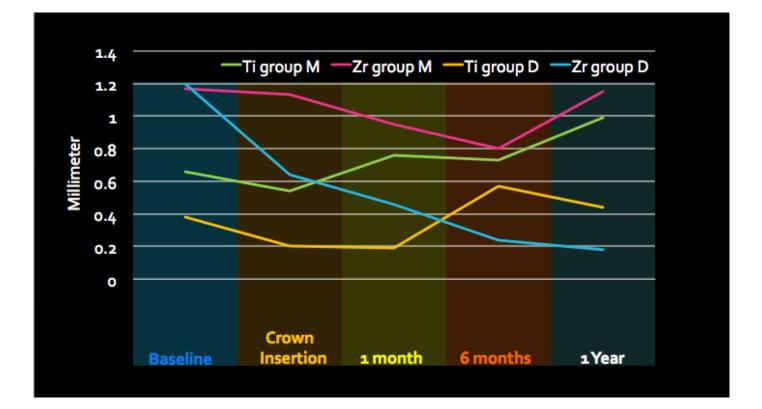


M: distance from the implant shoulder to first bone-to-implant mesially.

D: distance from the implant shoulder to first bone-to-implant distally.

Figure 6: Vertical bone changes (marginal bone)

Marginal bone changes occurred around the implant-prosthetic complex on the mesial and distal sites in both groups throughout the study time points.



M: marginal bone of the mesial adjacent tooth.

D: marginal bone of the distal adjacent tooth

Figure 7: Prevalence of the bacterial species around the adjacent teeth at the initial visit.

The graph represents the percentages of detection frequency levels for 40 bacterial species in both groups. Bacteria were grouped by their association into complexes as presented in Table 1.

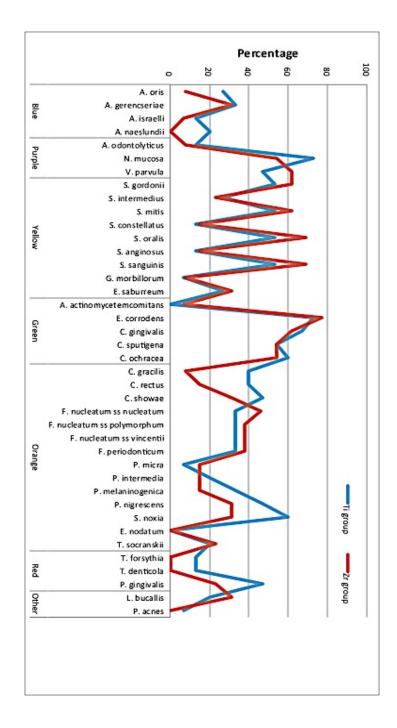


Figure 8: Prevalence of the bacterial species around the adjacent teeth at the final visit.

The graph represents the percentages of detection frequency levels for 40 bacterial species in both groups. Bacteria were grouped by their association into complexes as presented in Table 1.

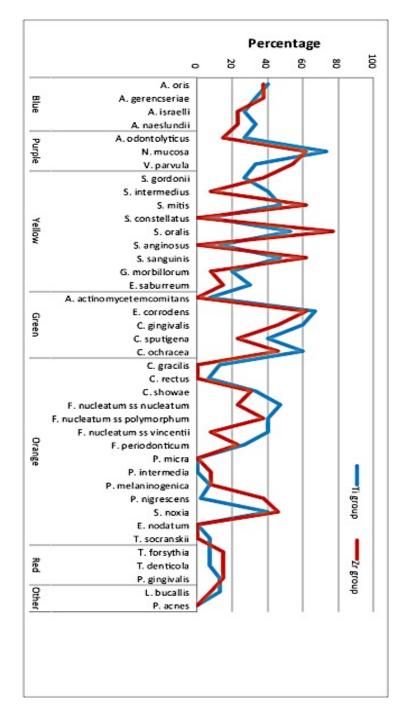


Figure 9: Prevalence of the bacterial species around the different abutments at the final visit.

The graph represents the graph represents the percentages of detection frequency level for 40 bacterial species in both groups. *Indicates significant statistical differences (p<0.00128). Bacteria were grouped by their association into complexes as presented in Table 1.

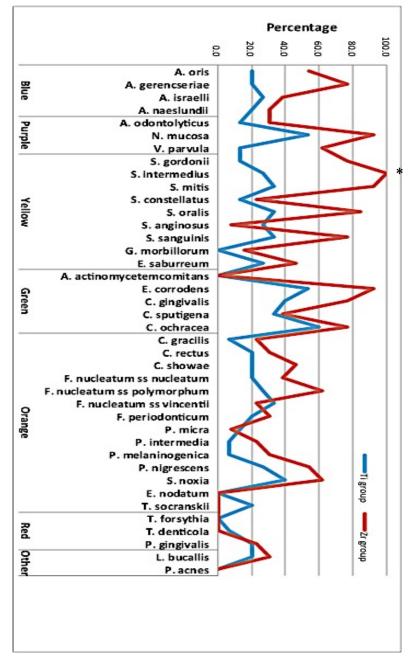


Figure 10: Percentage of DNA probes in the samples collected from the different abutment sites at the final visits.

The graph represents the percentages of DNA probes in the samples for 40 bacterial species in both groups. *Indicates significant statistical differences (p<0.00128). Bacteria were grouped by their association into complexes as presented in Table 1.

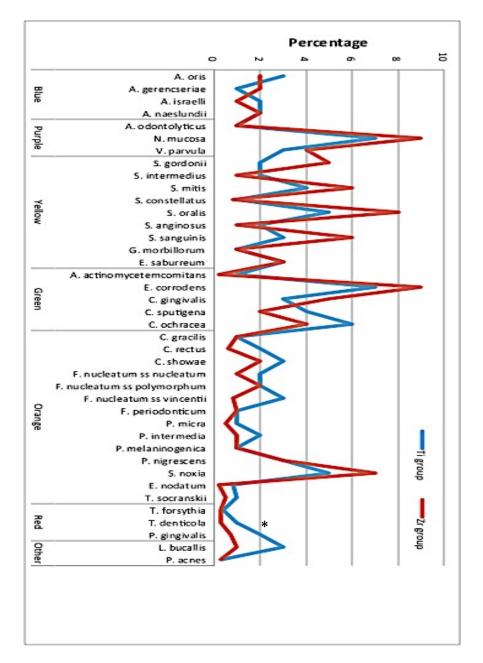
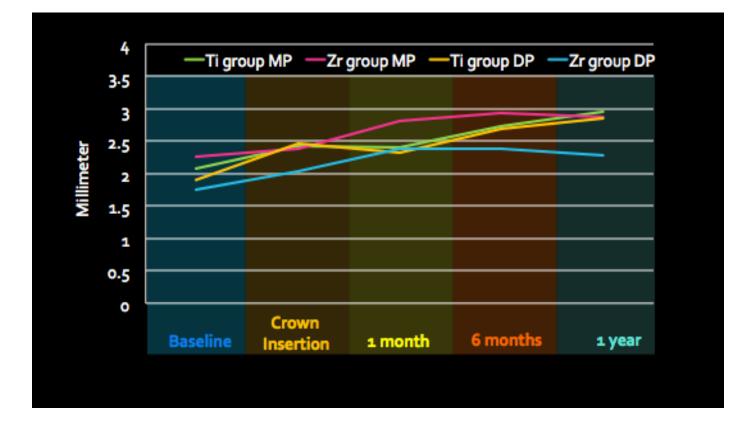


Figure 11: Papilla height around the implant crown

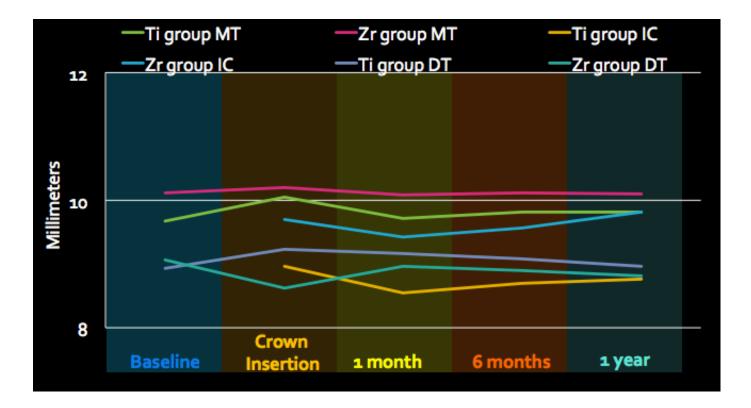
The graph represents the changes in the papilla height occurred around the implant-prosthetic complex on the mesial (MP) and distal (DP) sites in both groups throughout the study time points.



MP: mesial papilla. DP: distal papilla.

Figure 12: Clinical crown height

The graph represents the changes in the clinical crown height occurred at the implant crown (IC), the mesial adjacent tooth (MT), and the distal adjacent tooth (DT) in both groups throughout the study time points.



MT: mesial tooth. IC: implant crown. DT: distal tooth.

Figure 13: Patients' self-esteem

The graph represents the percentage patients scored regarding their self-esteem before and after treatment and throughout the follow-up visits in both groups.

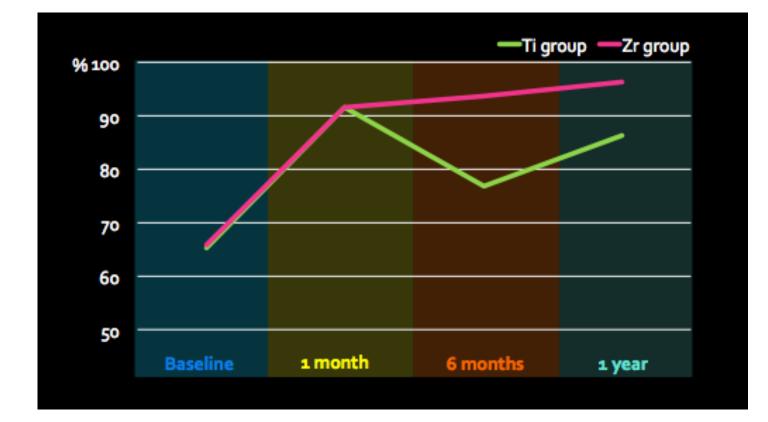


Figure 14: Patients' esthetic evaluations (subjective outcomes)

The graph represents the percentage of the expected esthetic outcomes the patients scored at the baseline visit and compares it with the accomplished esthetic outcome at the remaining time points in both groups.

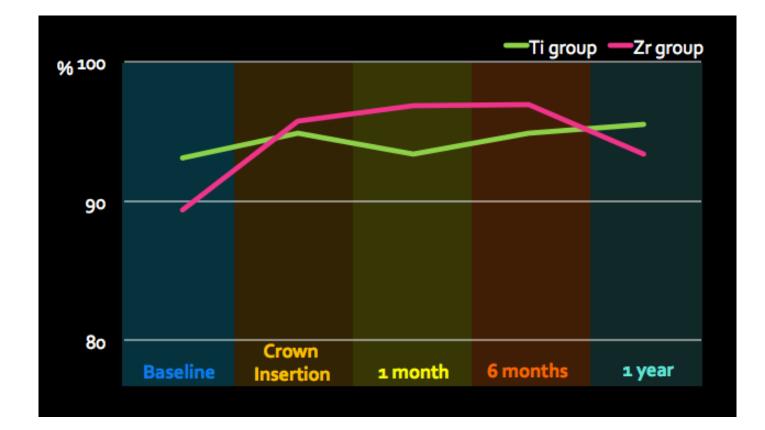


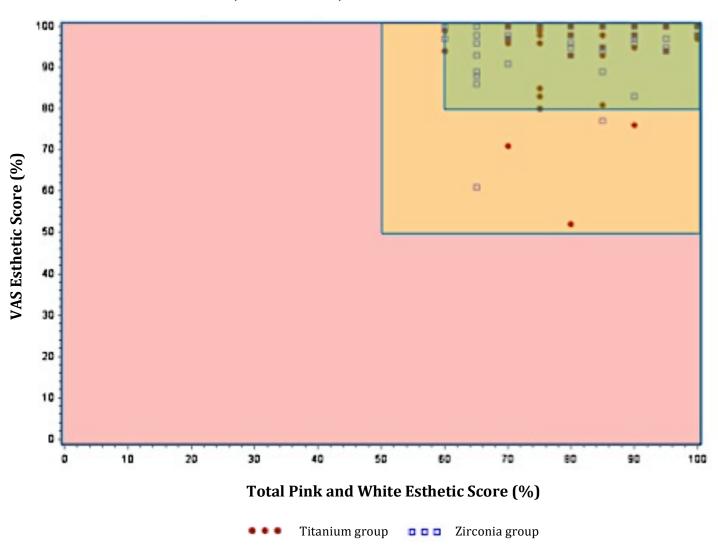
Figure 15: Patients' overall treatment satisfaction

The graph represents the percentage of overall treatment satisfaction the patients scored during different time points in both groups.



Figure 16: Subjective and objective esthetic classification (SOE)

Three categories are shown within the plot: satisfactory (green color), marginal (yellow color), and unsatisfactory (pink color).



Subjective and Objective Esthetic Classification

Table 1: Different bacterial species analyzed by DNA checkerboardhybridization

Different bacterial complexes have been described for subgingival samples and the various complexes were associated with different colors Socransky et al. [32] as listed in this table. The bacteria in yellow, blue, purple, green complexes are generally associated with gingival healthy, while the orange and red complex bacteria more frequently associated with periodontitis. A similar species distribution was described for peri-implant sites as in the current investigation of Shibli et al. [33].

Streptococcus	Actinomyces oris	Campylobacter rectus	Fusobacterium
anginosus			nucleatum ss
			polymorphum
Streptococcus	Actinomyces	Campylobacter	Fusobacterium
constellatus	gerencseriae	showae	nucleatum ss
			nucleatum
Streptococcus mitis	Actinomyces naeslundii	Campylobacter gracilis	Eubacterium nodatum
Streptococcus	Actinomyces israelii	Prevotella nigrescens	Fusobacterium
sanguinis			periodonticum
Streptococcus oralis	Veillonella parvula	Prevotella intermedia	Parvimonas micra
Streptococcus	Actinomyces	Fusobacterium	Bacteroides
intermedius	odontolyticus	nucleatum ss. vincentii	melaninogenica
Streptococcus gordonii	Neisseria mucosa	Selenomonas noxia	Treponema socranskii
Gemella morbillorum	Eikenella corrodens	Capnocytophaga	Tannerella forsythia
		ochracea	
Eubacterium	Aggregatibacter	Leptotrichia buccalis	Treponema denticola
saburreum	actinomycetemcomitans		
Capnocytophaga	Capnocytophaga	Propionibacterium	Porphyromonas
sputigena	gingivalis	acnes	gingivalis

Table 2: Biological parameters

Plaque scores, bleeding scores, width of keratinized mucosa, and probing depth of Ti and Zr groups accounting for overall follow-up period.

Variable		ening		eline		nsertion		onth		onths	1 y	Р	
		- 0									5		value
	Ti	Zr	Ti	Zr	Ti	Zr	Ti	Zr	Ti	Zr	Ti	Zr	
Teeth plaque score	2.30	2.88	2.67	2.07	2.52	2.00	2.04	1.85	1.65	1.68	2.27	1.33	0.32
	±0.99	±1.47	±1.16	±1.01	±1.30	±1.35	±1.27	±0.85	±0.77	±0.81	1.58	±1.01	
Implant plaque	NA	NA	1.00	0.64	1.00	2.08	1.31	1.92	1.31	1.45	0.92	1.42	0.09
score			±1.41	±1.22	±1.31	±1.71	±1.25	±1.44	±0.95	±0.93	±1.19	±1.31	
Teeth bleeding	0.31	0.15	0.30	0.61	0.57	0.08	0.35	0.23	0.31	0.09	0.77	0.33	0.053
score	±0.48	±0.24	±0.56	±0.59	±0.80	±0.19	±0.52	±0.39	±0.48	±0.20	±0.60	±0.39	
Implant bleeding	NA	NA	0	0.21	0.33	0.31	0.23	0.15	0	0.09	0.48	0.17	0.92
score				±0.58	±0.62	±0.63	±0.60	±0.38		±0.30	±0.88	±0.39	
Keratinized Tissue Width:													
Mesial tooth	NA	NA	5.00	4.57	5.20	4.93	6.62	5.07	6.53	5.64	6.69	5.50	0.14
			±1.36	±1.60	±1.52	±1.82	2.26	±1.54	±1.71	±1.86	±2.14	±1.31	-
Implant	NA	NA	5.87	5.50	5.80	5.14	6.38	4.86	6.31	5.36	6.46	5.17	0.09
-			±1.68	±1.83	±1.78	±1.61	±1.85	±1.23	±1.38	±1.36	±2.26	±1.34	
Distal tooth	NA	NA	4.46	4.64	4.53	5.00	5.77	5.43	5.92	5.63	6.23	5.83	0.94
			±1.56	±1.65	±1.60	±2.04	±1.53	±1.40	±2.14	±1.69	±2.42	±1.59	0.51
Probing depth		-	=1.00	_1.00	=1.00		=1.00	=1.10	-2.11	=1.0 9		=1.0 7	<u> </u>
i i obilig deptil													
Mesial tooth (mesio-	NA	NA	1.93	2.00	2.33	2.21	2.23	2.29	2.08	2.00	2.31	2.25	0.89
buccal)			±0.26	±0.78	±0.49	±0.89	±0.44	±0.73	±0.64	±0.77	±0.63	±0.75	
Mesial tooth (mid-	NA	NA	1.33	1.36	1.53	1.43	1.62	1.43	1.31	1.36	1.23	1.42	0.91
buccal)	1111		±0.49	±0.50	±0.52	±0.51	±0.65	±0.65	±0.48	±0.50	±0.44	±0.51	0.71
Mesial tooth (disto-	NA	NA	2.13	1.93	2.13	1.93	2.38	2.29	2.31	1.91	2.15	2.17	0.23
buccal)	INA	NA	±0.64	±0.73	±0.52	±0.62	±0.65	±0.61	±0.48	±0.83	±0.38	±0.83	0.25
													0.001
Mesial tooth (mesio-	NA	NA	2.20	1.93	2.00	1.71	2.07	1.86	2.08	1.64	2.38	2.00	0.02*
lingual)			±0.56	±0.62	±0.53	±0.61	±0.49	±0.36	±0.64	±0.67	±0.51	±0.60	
Mesial tooth (mid-	NA	NA	1.67	1.43	1.47	1.43	1.62	1.57	1.23	1.27	1.54	1.50	0.45
lingual)			±0.49	±0.51	±0.52	±0.51	±0.51	±0.51	±0.44	±0.47	±0.52	±0.52	
Mesial tooth (disto-	NA	NA	2.20	2.00	2.07	1.64	2.08	1.71	1.85	1.55	2.23	1.92	0.051
lingual)			±0.56	±0.68	0.46	±0.63	±0.49	±0.61	±0.55	±0.82	±0.44	±0.51	
Distal tooth (mesio-	NA	NA	1.87	2.00	2.40	2.07	2.46	2.21	2.31	1.91	2.38	2.25	0.23
buccal)			±0.52	±0.78	0.63	±0.83	±0.52	±0.58	±0.48	±0.70	±0.51	±0.75	
Distal tooth (mid-	NA	NA	1.33	1.36	1.40	1.57	1.62	1.36	1.07	1.18	1.23	1.50	0.79
buccal)	1111	1111	±0.49	±0.50	±0.51	±0.65	±0.61	±0.50	±0.28	±0.40	±0.44	±0.52	0.7 5
-	NI A	NA											0.10
Distal tooth (disto- buccal)	NA	NA	2.13	2.29	2.40	2.07	2.38	2.14	2.54	2.18	2.31	1.92	0.18
-			±0.52	±0.83	0.51	±0.83	±0.65	±0.53	±0.52	±0.87	±0.63	±0.51	
Distal tooth (mesio-	NA	NA	1.93	2.00	2.13	1.71	1.92	1.79	1.92	1.73	2.23	2.00	0.23
lingual)			±0.46	±0.68	±0.52	±0.61	±0.49	±0.58	±0.49	±0.79	±0.44	±0.74	
Distal tooth (mid-	NA	NA	1.53	1.43	1.40	1.57	1.38	1.50	1.31	1.64	1.23	1.50	0.26
lingual)			±0.52	±0.51	±0.51	±0.65	±0.51	±0.65	±0.48	±0.81	±0.44	±0.52	
Distal tooth (disto-	NA	NA	2.13	2.14	2.07	2.07	1.85	2.14	2.00	1.82	2.15	2.08	0.92
lingual)			±0.35	±0.86	±0.46	±0.73	±0.38	±0.53	±0.41	±0.75	±0.38	±0.51	
	D-1	a are pres											

Data are presented as mean ± standard deviation. Ti: titanium group. Zr: zirconia group. * Indicates significant statistical differences (p < 0.05)

Table 3: Esthetic parameters (objective outcomes)

Mean of PES and WES scores (according to Belser et al. 2009) between groups accounting for overall follow-up period. Data is presented as mean ± standard deviation. Ti: titanium group. Zr: zirconia group. PES: pink esthetic score. WES: white esthetic score.

Variable	Crown I	nsertion	1 M	onth	6 Mc	onths	1 y	ear	P value
	Ti	Zr	Ti	Zr	Ti	Zr	Ti	Zr	
Pink esthetic score:									
Mesial papilla	1.25	1.36	1.62	1.64	1.54	1.63	1.38	1.58	0.52
	±0.62	±0.50	±0.51	±0.50	±0.66	±0.50	±0.51	±0.67	
Distal papilla	1.42	1.29	1.62	1.36	1.69	1.36	1.69	1.25	0.11
	±0.67	±0.47	±0.51	±0.50	±0.48	±0.50	±0.48	±0.45	
Curvature of facial mucosa	1.58	1.50	1.46	1.43	1.77	1.54	1.77	1.58	0.22
	±0.51	±0.52	±0.52	±0.51	±0.44	±0.52	±0.44	±0.51	
Level of facial mucosa	1.50	1.50	1.77	1.57	1.92	1.54	1.92	1.67	0.21
	±0.52	±0.52	±0.44	±0.51	±0.28	±0.52	±0.28	±0.49	
Root convexity, soft tissue	1.33	1.50	1.62	1.36	1.38	1.27	1.62	1.50	0.58
color and texture	±0.49	±0.52	±0.51	±050	±0.51	±0.47	±0.51	±0.52	
Total PES score	7.08	7.14	8.08	7.36	8.31	7.36	8.38	7.78	0.26
	±1.51	±1.23	±1.04	±1.86	±1.18	±1.80	±1.19	±1.93	
White esthetic score:									
Tooth form	1.67 ±0.49	1.43 ±0.51	1.85 ±0.38	1.64 ±0.50	1.92 ±0.28	1.36 ±0.50	1.92 ±0.28	1.67 ±0.49	0.09
Tooth volume/outline	1.58	1.50	1.77	1.50	1.92	1.45	1.92	1.67	0.09
,	±0.51	±0.52	±0.44	±0.52	±0.28	±0.52	±0.28	±0.49	
Color	1.33	1.64	1.54	1.64	1.38	1.45	1.38	1.58	0.22
	±0.49	±0.50	±0.52	±0.50	±0.51	±0.52	±0.51	±0.51	
Surface texture	1.92	2.00	2.00	2.00	1.77	2.00	1.85	2.00	0.055
	±0.29	±0	±0	±0	±0.44	±0	±0.38	±0	
Translucency	1.50	1.86	1.77	1.71	1.62	1.73	1.54	1.83	0.21
	±0.52	±0.36	±0.44	±0.47	±0.51	±0.47	±0.52	±0.39	
Total WES score	8.00	8.43	8.92	8.50	8.62	8.00	8.62	8.75	0.97
	±1.21	±1.09	±0.95	±1.34	±1.39	±1.18	±1.12	±1.29	
Total PESWES score	15.08	15.57	17.00	15.86	16.92	15.36	17.00	16.33	0.46
	±1.93	±1.91	±1.78	±2.98	±2.06	±2.42	±1.87	±2.42	