Original Article

Microleakage Evaluation at Implant-Abutment Interface Using Radiotracer Technique

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Abstract

Objectives: Microbial leakage through the implant-abutment (I-A) interface results in bacterial colonization in two-piece implants. The aim of this study was to compare microleakage rates in three types of Replace abutments namely Snappy, GoldAdapt, and customized ceramic using radiotracing.

Materials and Methods: Three groups, one for each abutment type, of five implants and one positive and one negative control were considered (a total of 17 regular body implants). A torque of 35 N/cm was applied to the abutments. The samples were immersed in thallium 201 radioisotope solution for 24 hours to let the radiotracers leak through the I-A interface. Then, gamma photons received from the radiotracers were counted using a gamma counter device. In the next phase, cyclic fatigue loading process was applied followed by the same steps of immersion in the radioactive solution and photon counting.

Results: Rate of microleakage significantly increased (P≤0.05) in all three types of abutments (i.e. Snappy, GoldAdapt, and ceramic) after cyclic loading. No statistically significant differences were observed between abutment types after cyclic loading.

Conclusions: Microleakage significantly increases after cyclic loading in all three Replace abutments (GoldAdapt, Snappy, ceramic). Lowest microleakage before and after cyclic loading was observed in GoldAdapt followed by Snappy and ceramic.

Keywords: Dental Implants; Dental Implant-Abutment Design; Thallium Chloride

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INTRODUCTION

Various parameters influence the long-term success of dental implants such as microgaps, crestal bone loss and microleakage [1]. Bacterial leakage and presence of inflammatory cells at the implant-abutment (I-A) interface leads to bone loss around the existing microgap at the region of implant connection [2-4]. The rate of microleakage depends on several factors such as compatibility between components, torque amount, functional rocking, decrease of preload, screw loosening and component micro-movements [3,5-7]. Prevention of microbial leakage at the I-A interface is a major challenge in use of two-stage implant systems, which minimizes inflammatory reactions and maximizes bone stability at the implant neck [4]. The I-A interface

is often at or near the level of crestal alveolar bone and is usually located subgingivally, in most dental implant systems [8]. This area is an ideal site for plaque accumulation, which would allow microbial leakage. Several studies showed that even implant systems with high degree of compatibility of components could not completely prevent bacterial leakage and colonization [4,8].

The complications due to I-A misfit include increased microleakage, abutment rotation, screw loosening and decreased preload [9]. Transverse occlusal forces on prosthesis during function cause bending and micro-movement in the entire implant system, increased gap at the interface, and pumping effect between the interior surfaces of implant and tissues around it.

Several studies have shown bacterial microleakage at the I-A connection in different systems $[2,5-15]$.

Many techniques were suggested for evaluation of microleakage such as bacterial leakage model, fluid filtration, neutron activation, dyes, chemical tracers, scanning electron microscopy and radioactive tracers [3,10]. Most studies benefited from bacterial microleakage method using different size of bacteria, between 1-10 µm, for evaluation of I-A interface in dental implants [1-3,5,6,8,13,16,17]. The microgap size between the implant and prosthetic components ranges between 1 to 49 µm depending on the type of selected abutment [8,9,11,16]. The results are influenced by the type of bacteria, their size and their survival. The disinfection procedures can affect the results as well [3]. Lack of standardization in the methodology of bacterial leakage causes great variability in such studies and they cannot provide accurate information about the fluid leakage through the I-A microgap [3,16,18- 22]. There are several problems with bacterial leakage method, which may lead to false positive or false negative results [3]. The radiotracer method is quantitative, reproducible, and accurate. Also, due to the small size of radioisotopes, they have high degree of penetration. Anil et al, [8] studied microleakage of silicone liners and denture base with 45Ca radioisotope solution. Sarac et al, [10] measured microleakage of silicone liners and denture base with Thallium-201 chloride solution followed by gamma photon counting with a gamma camera. No previous study has evaluated microleakage of several abutment types at the I-A interface using radiotracing approach. Therefore, the aim of this study was to evaluate microleakage with thallium-201 in different abutments at the I-A interface and assess the influence of cyclic loading on microleakage. The null hypothesis was that there would be no significant difference in microleakage at the I-A interface in the three abutment groups before or after cyclic loading.

MATERIALS AND METHODS

In this study, 17 Replace implants (Nobel Biocare, Gotegerg, Sweden) and 17 abutments were used. The implants had 13 mm height and 4.3 mm diameter with regular platform. They were assigned to three groups of five samples. The following abutments were considered for each group: Snappy abutment (Nobel Biocare, Goteberg, Sweden) with 5 mm height, GoldAdapt engaging abutment (Nobel Biocare, Goteberg, Sweden) for the second group, and Zirconia abutment. Two implants with Snappy abutment with 0.5 mm collar, one for positive and one for negative control were also used.

Fabrication of ceramic samples

Zirconia blocks, machine aided design/ machine aided manufacturing (MAD/MAM) method and Zirkonzahn technology were used according to the instructions of the manufacturer. Snappy abutments were used as blueprint to build ceramic samples. Samples were built using 10 special burs and plumb line (for correct angulation of bur). The samples were immersed in A3 color liquid for five seconds. They were dried using an infrared lamp. Finally, the samples were placed in a furnace at 1500° C for eight hours, according to the manufacturer's instruction, to reach the desired stiffness (Fig. 1). Fabrication of ceramic abutment closely similar to Snappy abutment is of high importance especially at the interface area.

Fabrication of GoldAdapt samples

Snappy abutments were again used as blueprint in this case. GoldAdapt abutments are a subset of cast-to abutments. In terms of connection, they

Fig. 1: The fabrication process of customized ceramic abutment, (A) attaching Snappy abutment to Zirkonzahn machine as a model, (B) ceramic samples

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Fig. 2: Fabrication process of GoldAdapt samples, (A) wax up, (B) milling procedure, (C) GoldAdapt samples

are exactly the same as Snappy abutments. In order to maximize the precision and reliability of the results, the shapes of GoldAdapt and Snappy abutments were made as similar as possible. GoldAdapt abutments were mounted on implant analog and set in plaster. Then plastic sheaths of abutments were cut by a disc to proper vertical and horizontal extents. Using the inlay wax (Kerr, California, United States) and index prepared from Snappy abutment, an approximate contour of Snappy abutment was waxed up. The abutment screw was taken off and the model was removed from implant analog by removal tool. Following the instruction of the manufacturer, Carbon-free plaster and phosphate-bonded investment (Ceramvest, Protechno, Kerken, Germany) were used to build the mold. Castings were made using a low-fusing type III high gold (Degudent U, DeguDent GmbH, Rodenbacher, Germany) alloy. The casting temperature was $2350^{\circ}F$ (~1288 $^{\circ}C$) and burnout temperature was $1500^{\circ}F$ (~816 $^{\circ}C$) according to instructions of the manufacturer. To make GoldAdapt abutment further similar to the Snappy abutment, especially at the slots, a milling machine was used (Fig. 2).

Direct contact of beating arm with abutments, especially ceramic ones, during cyclic loading may result in cracking or breakage of abutments. Crowns with 45o angle were made using base metal alloy (Wironit; Bego, Bremen, Germany) to prevent direct impact on abutments [11]. Crowns were formed in a similar way and horizontal handles were attached to facilitate removing them after cyclic loading.

Microleakage assessment and cyclic loading

The abutments in all three groups were tightened using an electronic torque meter with the manufacturer's recommended torque of 35 N/cm. Then, abutments and implants were placed in putty (Speedex, Silicone Impression Material, Coltene/ Whaledent, Langenau, Germany) up to 1 mm from the I-A interface and the junction of putty and samples was covered with cyanoacrylate glue using a microbrush. This was done to minimize radiotracer's adherence to the outer surfaces of the abutment and implant, and to reduce errors during gamma ray counting. A great deal of attention was paid to make sure that the glue did not penetrate into the I-A interface while putty junction was completely sealed by applying as little glue as possible. The negative control samples were placed entirely inside putty but no putty was used for the positive controls (Fig. 3). The positive controls were designed to measure maximum radioisotope adherence to the samples and the negative controls were designed to

Fig. 3: Samples mounted in putty for measuring microleakage, (A) implant and abutment, (B) implant and abutment covered with putty (negative control), (C) applying adhesive with a brush

measure the minimum radioisotope adherence to the samples.

The samples were immersed in Thalliumchloride-201 radiotracer solution of 2 mCi (millicurie) in 500 ml water for 24 hours. Then the samples were removed, dried and putty was putty, close attention was paid to make sure the detached from the samples. When detaching radioactive contamination did not transfer from one sample to another.

Samples were placed in the same position in gamma ray counting container. A gamma counter device (Kontron, Gammamatic, Upplands Väsby, Sweden) with adjustment for Thallium-201 gamma (77 keV) and an energy window of 15% [11] were used to count gamma photons during a time interval of one minute. All measured values were recorded as gamma photon count per minute (cpm). Samples were shielded in lead for 12 days to prevent radioactive contamination.

At first, the implants were wrapped in thin layers of lead of radiographic films. This was done to prevent acrylic resin from remaining in the implant threats and to facilitate detachment of acrylic from implants after cyclic loading. Samples were placed in cyclic loading jig using a surveyor to make sure that the force was applied at the same direction as that of the implant abutment. In this situation, autopolymerized acrylic resin (Luxatemp; DGM, Hamburg, Germany) was placed inside the mold up to 1 mm below Interface. All samples were subjected to cyclic loading (Chewing simulator, S-D mechatronic, Feldkirchen-Westerham, Germany) for 500,000 cycles with a frequency of 2 Hz and force of 75 N. The end point of the force applying lever was round and it was adjusted on a 45-degree slope on the crown.

Samples were removed from acrylic blocks after cyclic loading. The same as before, preparation steps were performed for samples and they were placed in putty. Microleakage test was performed exactly similar to that of the first phase. There

were negative and positive controls similar to the first phase.

Repeated measures ANOVA was used to evaluate the differences in microleakage between the two groups before and after cyclic loading. P≤0.05 was considered statistically significant.

RESULTS

Two of the ceramic samples broke in the abutment region (Fig. 4). One-way ANOVA determined that there was no statistically significant difference among the three groups before and after cyclic loading (P>0.05). Repeated measure ANOVA was used for comparison of microleakage and detorques value. The different groups of abutments were considered as a Between Subject Comparison. The microleakage and detorque value before and after cyclic loading were considered as a Repeated Factor. The interaction effect was not significant ($P=0.678$). The repeated measure ANOVA was 0.015, the report between subject comparisons of this model was 0.382.

However, there was no statistically significant difference among the groups in the magnitude of increase in microleakage (P>0.05). Before cyclic loading, GoldAdapt abutments showed the lowest microleakage followed by Snappy abutments; the ceramic abutments showed the highest microleakage. However, these differences were not statistically significant (P=0.273).

Fig. 4: Two broken ceramic samples during cyclic loading process, the fractures were on the walls not connections

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After cyclic loading, GoldAdapt abutments showed the least amount of microleakage followed by Snappy and ceramic abutments. However, these differences were not statistically significant either (P=0.678). The mean and standard deviation values in the two groups are shown in Table 1.

DISCUSSION

At present, different implant systems are available with different connections, improved quality and special surface characteristics to minimize implant failure due to I-A interface misfit. However, there has been no study to assess and compare the microleakage of GoldAdapt, Snappy and customized ceramic abutments with radioisotope before and after cyclic loading. The selected cyclic loading protocol simulated about 18-24 months of clinical service [20].

The criteria for selection of the measurement method for microleakage evaluation usually include simplicity of use, precision, and size of gap. In many studies, dye penetration technique or bacterial leakage model at the I-A interface have been used, each with several complications [15]. Radioisotopes, due to their small size, penetrate more than pigments. Based on a study conducted by Charlton and Moore [15], radioisotopes show higher level of microleakage compared to other procedures. Gamma counters, which are classified among the nuclear medicine non-imaging detectors, have a very high sensitivity and specificity. The gamma photons counted by these devices can be a precise representative of the actual gamma ray emitted from the radioactive material; therefore, they are used in precise quantitative measurements [21]. It is reported that using radioisotopes provides more precise information about the amount of microleakage since the size of radioisotope particles is about 40 nm while the smallest pigment particle is 120 nm and the size of bacteria is in the range of 50 to 1000 nm [21].

Table 1: The mean and standard deviation values of micro-leakage before and after cyclic loading (CL).

Abutment Groups		Mean (cpm)	Standard Deviation
GoldAdapt	Before CL	22396.20	16124.54
	After CL	42227.00	15290.58
Snappy	Before CL	35639.20	11367.85
	After CL	55579.20	27133.62
Zirconia	Before CL	37909.00	16304.46
	After CL	59888.40	29332.36

Radioisotope offers a precise method, which is relatively inexpensive and reproducible. It provides the opportunity to measure microleakage quantitatively. The samples in this method are totally recoverable and after the test they can be used for other tests. Since the radioisotope method is very sensitive, shape and surface texture of a sample can partially influence the level of adherence of radioisotopes to the sample. Therefore, it is crucial to make the samples as similar as possible to each other.

Washing the samples with water might result in removing the radioisotopes from the I-A interface, so the operator must pay attention to this issue; otherwise, it may cause inaccuracies in results [22].

do Nascimento et al, [6] stated that cast-to and pre-machined abutments had lower leakage when the manufacturer's instructions were followed. This result was in line with our findings. However, in the study by do Nascimento et al, [6] the samples did not undergo cyclic loading while in our study the samples were evaluated before and after cyclic loading, which better simulated the clinical oral environment.

Hjerppe et al, [19] compared the load-bearing capacity of custom-made and prefabricated zirconia abutments. Their conclusion was that the custom-made and prefabricated abutments were at the same level from the failure load point of view. However, marginal adaptation of custommade abutments was not as good as those of prefabricated ones [19]. These results were in

accordance with our findings showing that the largest amount of microleakage belonged to custom-made ceramic group although this difference was not statistically significant. Hjerppe et al, [19] only used static loading in their study; if they had used cyclic loading their results could have been different. Also, they used implant analog, which would behave differently compared to titanium. In their study, the custommade ceramic abutments showed higher microleakage, which indicated greater misfit at the I-A connection in this type of abutment [19]. In our study, two ceramic samples broke during cyclic loading (at the wall of abutment above the connection) probably due to the brittleness of ceramic. This is one of the disadvantages of ceramic abutments in comparison with metal abutments. The above-mentioned failure shows the ceramic abutment's lower resistance against loading compared to the other two abutment types [23].

The metal abutments have excellent survival rates due to their physical properties [23]. Metals are ductile, which results in their higher resistance against small defects and cracks. On the other hand, ceramics are brittle and as a result they cannot tolerate tensile forces and easily crack [23]. In our study, this difference was clearly observed. Moreover, due to shrinkage during the sintering process of ceramics, the wall thickness of custom-made ceramic abutments becomes a little thinner than that of snappy abutments, which increases the likelihood of failure in ceramic abutments. This was one of the challenges of this study to keep the custom-made ceramic abutments within the range of the target abutment after contraction when using MAD/ MAM method.

It is worth to mention that there is no study concerning the level of microleakage of MAD/ MAM ceramic abutments in comparison with factory-prefabricated titanium abutments. In the current study, this comparison was performed and as mentioned, the leakage level of ceramic abutments was found to be more than that of other groups with no statistical significance.

In the current study, microleakage level of GoldAdapt abutments before and after cyclic loading was lower than that of other groups with no statistical significance. Significant differences might have been achieved if a larger sample size had been used. Therefore, more studies with larger sample sizes are required in this respect.

The results of the current study supported our null hypothesis, i.e., there would be no significant difference in microleakage at the I-A interface before and after cyclic loading among the study groups. These concepts need to be addressed in future studies with larger samples, more cycles of cyclic loading, assessment of external abutments and other types of connections.

CONCLUSION

Within the limitations of this study, it can be concluded that:

1- Radiotracer technique is a precise and sensitive method for evaluation of microleakage in I-A interface.

2- Microleakage increases significantly after cyclic loading in all three Replace abutments (GoldAdapt, Snappy, ceramic).

3- Lowest microleakage before and after cyclic loading was observed in GoldAdapt followed by Snappy and ceramic. However, this order was not statistically significant.

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