Histomorphometry and Stability Analysis of Loaded Implants with two Different Surface Conditions in Beagle Dogs

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Despite an improved bone reactions of Mg-incorporated implants in the animals, little yet has been carried out by the experimental investigations in functional loading conditions. This study investigated the clinical and histologic parameters of osseointegrated Mg-incorporated implants in delayed loading conditions. A total of 36 solid screw implants (diameter 3.75 mm, length 10mm) were placed in the mandibles of 6 beagle dogs. Test groups included 18 Mg-incorporated implants. Turned titanium Implants served as control. Gold crowns were inserted 3 months. Radiographic assessments and stabilitytests were performed at the time of fixture installation, 2nd stage surgery, 1 and 3 months after loading. Histological observations and morphometrical measurements were also performed. Of 36 implants, 32 displayed no discernible mobility, corresponding to successful clinical function. There was no statistically significant difference between test implants and controls in marginal bone levels (p=0.413) and RFA values. The mean BIC % in the Mg-implants was 54.4 ± 20.2%. The mean BIC % in the turned implant was 48.9 ± 8.0%. These differences between the Mg-implant and control implant were not statistically significant (P=0.264). In the limitation of this study, bone-to-implant contact (BIC) and bone area of Mg-incorporated oxidized implant were similar to machine-turned implant. The stability analysis showed no significantly different ISQ values and marginal bone loss between two groups. Considering time-dependent bone responses of Mg-implant, it seems that Mg-implants enhanced bone responses in early loading conditions and osseointegrated similarly to cp Ti implants in delayed loading conditions. However, further investigations are necessary to obtain long-term bone response of the Mg-implant in human.

Key words: biochemical bonding, Mg-incorporated implant, delayed loading, RFA, BIC

(J.K.A.of Stomatognathic Function and Occlusion 2008;24(4):337-349)

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Received: March 18, 2008, Last Revision: December 03, 2008, Accepted: December 25, 2008

INTRODUCTION

Titanium and its alloy have been widely used for dental implants. Despite the successful clinical performance of cp titanium implants, it is still lacking of fundamental mechanism of osseointegration.[1] The osseointegration for cp titanium implants was achieved by rigid fixation between bone and implants. The fixation by a bone ingrowth was considered as a biomechanical interlocking. Recently biochemical bonding was proposed as another way of osseointegration with chemically modified titanium implants.[2] Although it has tremendous potential, biochemical bonding aspects of osseointegration have not been fully defined.

Various chemical modifications were suggested for improvement of the tissue responses. One of the chemical modifications is an electrochemical oxidation method. Sul et al.[2-4] reported on the significance of ion incorporation in titanium oxides such as S, P, Ca, respectively. S-incorporated, P-incorporated or Ca-incorporated oxidized implants were compared to the machine-turned implants by removal torque test and histomorphometric analysis. These results showed significantly improved bone responses compared to control implants. Especially, surfaces with incorporated calcium ions showed significantly enhanced integration strength of bone to implants compared to S-incorporated or P-incorporated implants. Sul et al.[5] also investigated surface oxide chemistry to bone responses using Mg-incorporated oxidized implants. In their reports, the optimal surface properties of Mg-incorporated implants have been proposed. Those included approximately 9.3% atomic concentration of Mg, the mean oxide thickness of 3400 ㎛, and crystalline structure by anatase and rutile type etc.[6]

Incorporated Mg^{2+} and Ca^{2+} ions were known to play essential roles in the binding interactions of the integrin superfamily of cell-surface receptor and the ligand proteins such as fibronectin, vitronectin, fibrinogen, and some cell-cell adhesion receptors.[7] Sul et al.[5] observed the chemical shifts of binding energy of atoms. They suggested that Mg^{2+} ion movements and exchanges are important driving forces to promote the integrin-ligand interactions that subsequently result in improved bone response via biochemical bonding of Mg^{2+} ion. Parise et al.[8] found that platelet receptor GPⅡb-Ⅲa more strongly bound to fibronectin in the presence of Mg^{2+} ion than in the presence of Ca^{2+} ion. Zreiquat et al.[9] investigated the better effect of Mg^{2+} ion on the cell adhesion, integrin expression and activation of intracellular signaling molecules than Al_2O_3. Martin and Brown[10] investigated magnesium's role during synthetic hydroxyapatite formation in vitro and possible related effects during biomineralization. These researches have applied to interpretation of biochemical bonding mechanism of Mg-incorporated titanium implants.

Recently, Sul et al.[5] investigated the bone tissue responses to Mg-incorporated oxidized titanium implants and machine-turned titanium implants in the rabbit femur. They reported the results of the resonance frequency analyses and removal torque measurements of the Mg-incorporated implants. The results demonstrated significant improvements in implant integration with the bone comparing to machine-turned implants. In the previous study, Kim[11] evaluated histomorphometry and stability analysis of Mg-incorporated implants with beagle dogs in early loading conditions. As the results, Mg-incorporated implants demonstrated significantly more bone-to-implant contact in early loading conditions. Sul et al.[6] compared the speed and strength of osseointegration and osteoconductivity between Mg-incorporated implant, an oxidized commercially available TiUnite® implant and

Osseotite® implant in rabbit model. They suggested that surface chemistry facilitated more rapid and stronger osseointegration of the Mg-incorporated implants despite their minimal roughness compared to the moderately roughened TiUnite®. These reports provided positive evidences for the surface chemistry-mediated biochemical bonding of bioactive implants.

Although many studies showed improved bone responses of Mg-incorporated implants, little yet has been carried out by the experimental investigations under functional loading conditions. Moreover, because the focus is extremely placed on shortening a healing period, a research about how Mg-incorporated implants have an effect on the bone response after a sufficient healing period has not been investigated. The aim of present study was to compare bone response of Mg-incorporated oxidized implants with turned implants in beagle dogs under delayed loading condition.

MATERIAL AND METHODS

1. Implant design and preparation

Thirty six screw-type implants (ASTM Grade IV) with a pitch height of 0.6 mm, outer diameter of 3.75 mm, length of 10 mm were manufactured. In the present study, 2 groups of titanium implants were used: turned implants were used as a control group and electrochemically oxidized implants incorporating Mg (Mg-implants) in the surface titanium oxide layer were test group. The test Mg-implants were prepared using the micro arc oxidation (MAO) process at high anodic forming voltages and current densities at galvanostatic mode in a magnesium containing mixed electrolyte system. The electrochemical oxidation method employed was as described in Sul's studies.[12]

2. Surface characteristics of the controls and test Mg-implants

Chemical surface analysis was investigated by XPS (ESCALAP 250, VG Scientific Ltd). The XPS spectra was recorded using normal Al $K\alpha$ radiation (1486.8 eV) with a probing beam size of 200 μm. The outmost surface of the implants was etched with Ar ion with ion energy of 5 KeV and a beam current of 0.3 μA for 150 seconds, corresponding to 2 nm in thickness, resulting in removal of surface contaminants. The surface oxides of the controls and Mg-implants consisted mainly of TiO_2. XPS survey spectrum of the Mg-implants revealed the presence of the Mg elements, Ti, O, C and some traces like P and S (Fig. 1a, 1b). The relative atomic

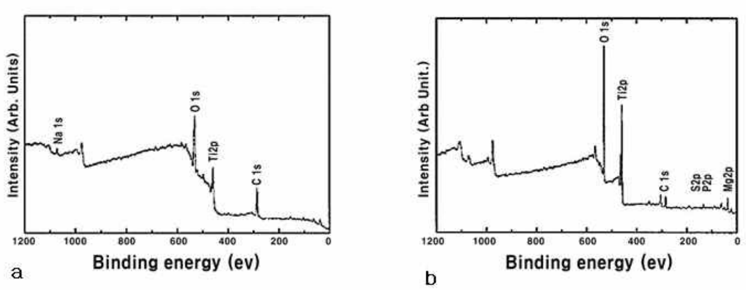

Fig. 1. XPS survey spectra. (a) turned implant, (b) Mg-implant.

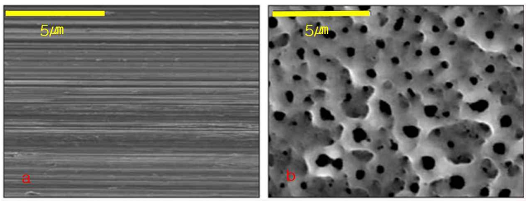

Fig. 2. SEM image.
(a) turned implant, (b) Mg-implant.

concentration of Mg was approximately 7.6% at the as-received surface and 9.3% after Ar+ sputter cleaning for 150 seconds. 15% of C at the as-received surface was rapidly decreased after sputter cleaning. This could indicate that the C is a surface contaminant. The mean surface porosity was 23.7%. The pore size was ≤ 1.5 ㎛ in diameter. The pore density (pore population/scanning area) was 3.25. The mean oxide thickness was 3.4 ㎛ in the test Mg-implants as measured with a scanning electron microscope (S-3000N; Hitachi Science System, Tokyo, Japan) (Fig. 2a, 2b) on a cross-section view and < 17 nm in the controls as measured with Auger Electron Spectroscopy (AES). The surface roughness measured with Optical Interferometry (MicroXam^{TM}) revealed Sa (arithmetic average height deviation) of 0.68 ㎛ (±0.2), Sdr (developed surface ratio, i.e., the ratio of the increment of the interfacial area of a surface over the sampling area) of 26.3% (±11.1), Sds (the number of summits of a unit sampling area) of 0.12 ㎛$^{-2}$ (±0.04) for the test Mg implants and Sa of 0.55 ㎛ (±0.21), Sdr of 10.6% (±3.9), Sds of 0.09 ㎛$^{-2}$ (±0.04) for the controls.

3. Experimental animals

This study was approved by the Animal Care Committee of the University of Kangnung, Korea. Six male beagle dogs, 24 months of age with mean body weight of 13kg, were used in this study. Large male beagle dogs were selected rather than smaller female species to ensure adequate alveolar ridge size and height for implant placement. All subjects were healthy and had no periodontal disease.

4. Surgical procedure

Removal of teeth in the mandible and implant placement was performed under sterile conditions and general anesthesia in an animal operating theater. Premedication included acepromazine IM (Sedaject; Samu Median Co., Yesan, Korea) and sadation was achieved by a combination of ketamine hydrochloride (Yuhan-Ketamine; Yuhan Co., Seoul, Korea) and xylazine hydrochloride (Rompun; Bayer Animal Health Co., Seoul, Korea). Initial dosage was 5 cc, and additional dosage 3 cc for maintaining anesthesia. Local infiltration was also performed with 2% lidocaine (Lidocaine HCl; Huons Co., Seoul, Korea).

Before removal of the teeth, bilateral impressions (Alginate Jeltrate; Dentsply International Inc, Milford DE, USA) of the mandible were made for fabrication of plaster casts and individual trays. Bilateral four premolars were extracted on each side with special care. The wound closure was carried out with Vicrly 4-0 (Ethicon GmbH, Norderstedt, Germany).

After a healing period of 3 months, implant placement performed atraumatically under sterile conditions. Crestal incision was made and a mucoperiosteal flap was reflected. With copious irrigation, implant site was sequentially enlarged using round bur, 2.0 mm twist drill, pilot drill, and 2.85-3.35 mm twist drill, countersinking drill according to the standard protocol (Nobel Biocare, Göteborg, Sweden). All the inserted implants anchored superior-cortical bone only and bucco-lingually about 1mm bone remained around the implants. A distance of approximately 7 mm between implant centers was maintained. All implants were placed so that the top of each was more or less flush with the alveolar crest after which flaps were repositioned and sutured with mattress and single interrupted 4-0 Gore-Tex sutures (Gore-Tex® Suture; W. L. Gore & Associates, Inc., Newark, USA). In each animal, 3 turned implants (control groups) were placed in one side of the mandible and 3 Mg-implants (test groups) in the other side with matching position.

Postoperatively, the animals were given antibiotics (Kanamycin-100; Samyang Pharma-Chem. Co., Seoul, Korea) and analgesics (Hipyrine; Samyang Pharma-Chem. Co., Seoul, Korea) for 7 days. The sutures were removed 10 days after surgery.

The animals were fed a soft diet for two weeks and were inspected in the post-operative days for signs of wound dehiscence or infection cautiously and later weekly to assess general health. Throughout the total period of the investigation, a systemic oral health care regimen was performed with three times of weekly tooth brushing with a soft brush and 0.2% chlorhexidine gluconate gel (USP, Maryland, U.S.A.).

5. Prosthetic loading

After 3 months for osseointegration, the surgical sites were exposed for healing abutment connection and impression taking. Non-rotational fixture level impression copings (Neoplant®; Neobiotech, Seoul, Korea) were connected to implants and secured with screws. Using individual tray and polyether impression material (Impregum; ESPE Co., MN, USA), pick-up impression was taken.

In 2 days after impression, Total 36 single gold crowns (type III gold, DM 76; Wooridongmyung Co., Seoul, Korea) were restored on the implants and intraoral adjustment was performed to eliminate any direct occlusal contact on closure in implanted areas. Absence of direct occlusal contacts between the maxillary teeth and the implant-supported restorations was verified with occlusal articulating paper (Accufilm Ⅱ; Parkell Inc, NY, USA). Gold crowns were subsequently screwed to the abutments with an tightening torque of 20 Ncm.

Twelve weeks after loading, all of the animals were sacrificed by induction of a deep anesthesia followed by intravenous overdose of sodium-pentobarbital and perfused with a fixative through the carotid arteries.

6. Specimen preparation and analysis

1) Radiographic analysis

During the experiment, the measured radiographic parameters are as follows: (1) presence or absence of peri-implant radiolucency, (2) measurement of marginal bone levels and calculation of changes over time.

Standardized periapical radiographs using a customized occlusal index fabricated by affixing a poly-vinyl siloxane putty impression material were taken. Thus, individual XCP could be attached to a portable long cone X-ray machine and parallel images were obtained.

Each radiograph was then computer digitized and analyzed for changes in bone levels (Adobe Photoshop 7.0.1 version, Seattle, USA). In the radiographs the distance between the most coronal part of the fixture (i.e., the abutment-fixture junction) and the most coronal bone judged to be in contact with the implant surface was determined at the mesial and distal aspects of each implant. The radiograph measurements were performed as bone level changed from implant placement to 3 months after loading.

2) Stability analysis

Implant stability was assessed by resonance frequency analysis (RFA, Osstell™; Integration Diagnostics Ltd., Göteborg, Sweden). Resonance frequency was measured by attaching the transducer (Type F1L5; Integration Diagnostics Ltd., Göteborg, Sweden) to the screw implant. The measurements were analyzed and implant stability quotient (ISQ) values at the time of fixture installation, 2nd stage surgery, 1 and 3 months after loading were compared.

3) Histomorphometric analysis

Specimens of the implants and surrounding tissues were removed en bloc from mandibles of all of the animals. The samples were fixed by immersion in 10% neutral buffered formalin solution (Accustain®; Sigma-Aldrich®, Inc., Steinheim, Germany) for 1 week and then dehydrated in ascending concentrations of ethanol, embedded in Technovit 7200Ⅱ VLC resin (Kulzer, Friedrichsdorf, Germany) and polymerized in a light polymerization unit (Exakt System, Exakt Apparatebbau, Norderstedt, Germerny). Transverse sections, 100㎛ in thickness, were taken along the long axis of the fixture in a mesiodistal direction with a band saw with a diamond blade (Exakt-Cutting Grinding System, Exakt Appratebau, Norderstedt, Germany). The final section was ground to approximately 20 ㎛ or less utilizing an Exakt Micro Grinder, and polished to an optical finish according to the cutting-grinding technique described by Donath and Breune.[13] Finally, all of the sections were stained with 1 % toluidine blue solution and were examined by light microscopy.

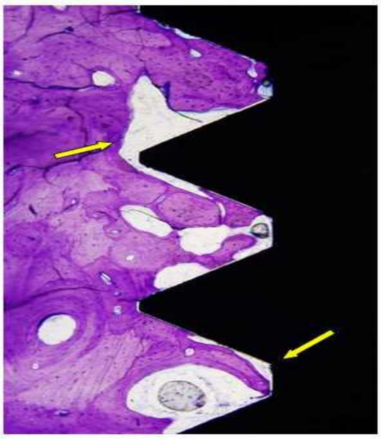

Fig. 3. Bone-to-implant contact (BIC, %) at the interface was measured to 7th thread of implant surface. In some areas, the bone actually did not contact with the thread (arrows).

Histomorphometric analyses were performed to get more information about the quality of the implant-tissue interface. The data were reported as percentage of bone-to-implant contact (BIC) (Fig. 3) and percentage of total mineralized bone tissue within the threads (referred to as "bone area"). First bone contact was measured from the distance between the abutment-fixture junction and the most coronal part of direct bone-to-implant contact. Digital photographs (DP70 microscope digital camera, Olympus America Inc., Melville, NY, America) were taken from all of the ground sections (magnification ×10) to permit morphometric assessment with microscope (BX-50 Fluorescence Microscope, Olympus, NY, America) connected to an IBM PC, equipped with DP controller computer software. Then the bone-to-implant contact and bone area measurements were made to 7th threads in the surface of each implant using Image J Program. For each implant, all measurements were repeated twice by one investigator.

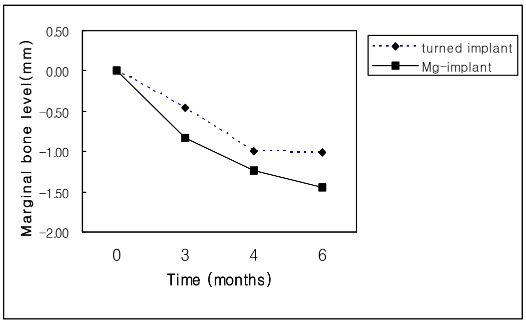

Fig. 4. Marginal bone level changes over time.

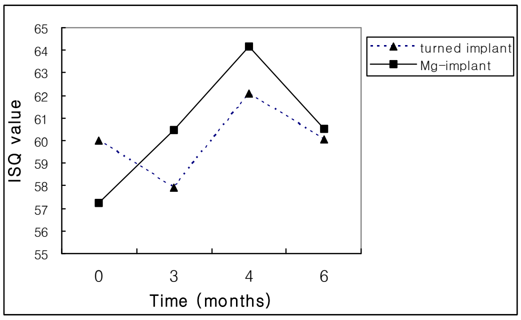

Fig. 5. ISQ Value changes over time.

7. Statistical analysis

Analysis of covariance (ANOVA) with repeated measurements was used to evaluate associations between the implant type and RFA values, implant type and the marginal bone levels over time. Statistical analyses of the histomorphometric measurements were performed using the Mann-Whitney U test. Statistically significance was considered at $P \leq 0.05$.

RESULTS

1. Clinical observations

Nine of 36 implants were lost. Four of the 18 turned implants and 4 of 18 Mg-implants failed to integrate due to sings of infection. Failure of 5 implants was caused by the post-surgical complications in one dog. The flap necrosis occurred and healing by secondary intention failed. The soft tissues around 3 of 36 implants showed recession with the presence of inflammation. After 3 months of loading, 1 Mg-implants showed the signs of failing implant and was excluded from the data. Any other implants did not show mobility and sings of peri-implant infection. Overall survival rate was 75%.

2. Radiographic analysis

Periapical radiolucencies were absent on radiographic examination throughout the study. The results from the radiographic measurements are showed in Fig. 4. The marginal bone loss following implant installation of test groups was slightly higher than control groups. During healing period, marginal bone loss was recorded about 0.5 mm at the control sites, and 0.8 mm at the Mg-implant sites. Both groups showed gradual bone loss. Comparing the bone levels between baseline and 3 months of loading, a statistically significant differences were found with the lapse of time, demonstrating clear time effect (P=0.000). Overall bone loss was 1.0 mm in the control group and 1.5 mm in the Mg-implant group. However, there was no statistical difference between the type of implant surface (P=0.413).

3. Stability analysis

The RF values varied as a function of time (Fig. 5). At baseline, mean ISQ value was 60.0 ± 4.8 in the control group and 57.2 ± 5.8 in the Mg-implant group. Mean ISQ was slightly decreased in the control group at the second stage surgery. Mean ISQ value in Mg-implant groups increased over time and was similar to the control groups after 3 months of loading (60.1 ± 9.1 vs 60.5 ± 7.9 for the control group and Mg-implant group, respectively). The statistical analysis showed that the differences between the implants at implant installation and at 3 months after loading (P=0.041). However, repeated measured ANOVA did not demonstrate the statistically significant difference between Mg-implants and controls through overall measuring time (P=0.684).

4. Histomorphometric analysis

The mean BIC value in the Mg-implants was 54.4 ± 20.2%. The mean BIC% in the turned implants was 48.9 ± 8.0% (Fig. 6). These differences were not statistically significant (P=0.264). Mean BIC in anterior installation site regardless of the implant surface was 37.9%. These were 54.7% and 61.8% in middle and posterior installation site, respectively. The mean BIC increased from the anterior site to the posterior site. These data showed a significant difference (P=0.038).

The results from histomorphometric analysis about bone area were 69.1 ± 8.6% in control implants and 58.9 ± 22.8% in test Mg-implants. Comparing the bone area inside the threads, the control implant group had more bone fill than the Mg-implants. However, there was not significantly difference (P=0.109).

First bone contact of Mg-implants and turned implants was 1.2 ± 0.6 mm, 1.0 ± 0.3 mm respectively in the histomorphometry. These were not significantly different (P=0.771).

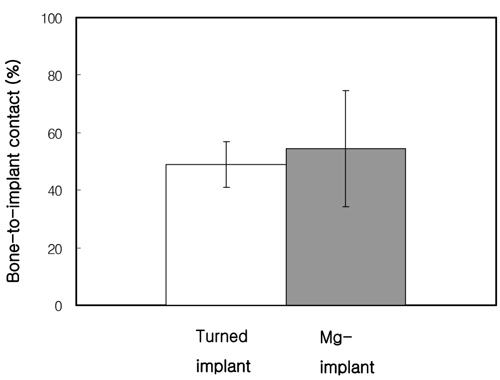

Fig. 6. Bone-to-implant contact (%).

DISCUSSION

This study was designed to evaluate the stability and bone response of Mg-incorporated oxidized implants under 3 months of delayed loading periods. While the previous study provided the effect of early loading condition applied to Mg-implants, given current loading protocols of 3 months, this study had a sufficient healing time. Comparison of the data between the 2 healing studies provides insight into time-dependent bone responses of Mg-implants.[11]

Five of 36 implants failed to osseointegrate in one dog after first surgery. It is well known that excessive surgical trauma and thermal injury may lead to osteonecrosis and result in fibrous encapsulation of the implant.[14] One Mg-implants failed after 3 months of loading. Five of 8 failed implants were placed at the most distal area. According to investigation of the present study, the narrow ridge shape and variable bone quality were most prominent in the anterior site adjacent to canine. The unfavorable conditions in anterior sites expected to us that failing frequencies of anterior implants area were higher than those of any other site. However, higher failure at the most distal area really might be related uncontrolled occlusal loading; transmission scheme of occlusal force in beagle dog differs in the human situation. Besides by exposure to functional loading without direct occlusion, the stress might be exerted with diet food on occlusal faces and through free-standing implants without splinting. This might affect a distinct magnitude of motion at most distal the implant-bone interface.

For peri-implant bone levels over time, a significant change was seen from the day of implant placement to 1-month after loading (1.0 mm in control groups, 1.2 mm in test groups). The largest changes occurred after first and second surgery and to a lesser degree after insertion of restorations. These results proposed that surgery and postsurgical healing events may be a greater contributor to bone level changes than actual loading. This was confirmed in a animal study by Cochran et al.[15] and clinical study by Pham et al.[16] This study showed that an average bone loss was 1.0 mm in turned implant and 1.5 mm in Mg-implant at the end of 3 months of loading. In the dog mandibles, Pilliar et al.[17] observed 1.2 mm loss of crestal bone after 6 months of loading with machined implants. Isidor[18] reported the bone loss in delayed loading implants within the range of 0.4 to 4 mm. In the previous study, an average bone loss was 1.5 mm in turned implant and 1.3 mm in Mg-implant in early loading condition. In this study, delayed loading implants had similar amount of bone loss to that of the early loading implants. On the other hand, Randow et al.[19] showed 0.8 mm bone loss of delayed loading was twice the amount of bone loss of early loading in clinical study. However, this difference may be associated with controlled and uncontrolled study by human and experimental animal trial.

The resonance frequency analysis has been suggested as a valuable method for non-destructive and quantitative assessment of stability of implant. It has been increasingly served as a sensitive and objective tool for clinical monitoring. This study showed no significant differences in ISQ values between turned implants and Mg-implants. Sul et al.[5] reported Mg-incorporated oxidized implant showed higher resonance frequency values when compared to the machine-turned control implants in rabbit experiment (67.4 in Mg-implants vs 64.0 in control implants). These differences may be related to loading protocol without direct occlusion. This study, in the loading environment, was provided by functional loading in beagle dogs whereas Sul's study with rabbits was not loaded functionally. Like in the result of failures of implant, adverse mechanics of free-standing implants compared with splinted implants and uncontrolled function in the oral sites could jeopardize stability of both implants. Moreover, the same geometric design between two groups might attributed to the result.

It is not possible in the present study to observe any significant difference in the percentages of bone to implant contacts between the test and control groups (BICs in the Mg-implants was about 54.4%, in the control implants about 48.9%). Comparison of the findings from this study with the previous study of BIC of Mg-implants improved presents similar data (In the previous study BIC in the Mg-implants was about 54.5%, in the control implants about 45.3%). In despite of similar data, few statistically significant differences were found between Mg-implants and turned implants. The reasons are as follows. First of all, standard deviation of Mg-implants is larger than control implants. Larger standard deviation may confuse statistical analysis. Moreover, because of the small sample size when dogs are considered as subjects for the longitudinal analysis (n=6) and for each time interval, statistical power, the ability of the experiment to detect a significant difference, is very low. Therefore, the lack of statistically significant differences between groups may the result of larger standard deviation of Mg-implants and the small sample size. Second, it is difficult in controlling oral hygiene of beagle dogs.

An adequate hygiene has always been considered an important requirement during the healing period.[20] Although consequent mechanical plaque control has been proposed even in animal trials,[21] plaque control in animal experiment is difficult in being carried out actually. When it occurs signs of infections during the healing period of the implants, intervention trial of animal study is more troublesome than human study. In this study, some Mg-implants showed inflammation accompanied with gingival recession despite of an effort to control hygiene. This might misinterpret the result of Mg-implant. In addition, the test implants were prepared up to implant top by micro arc oxidation method, whereas the controls had polished collar. These differences of surface roughness of the coronal part might be susceptible to the inflammation of Mg-implant. Third, it was thought to be the possibility of catch-up remodeling of Mg-implants. The previous study suggested chemical bonding by Mg played a important role in initial healing phase. However, a sufficient healing time and a long loading period might permit complete bone regeneration and counterbalance the effect on bone responses of Mg-implant in an early loading period by catch-up remodeling. Comparing cp Ti and HA-coated screws in the dog mandible, Evans et al.[22] did not find any significant differences in BIC between smooth and rough implant surfaces after functional loading for 6 months. Watzak et al.[23] showed no significant difference in BIC between smooth surface cp Ti screws and rough surface grit-blasted and acid etched screws after 18 months of functional loading in the mandible. They suggested that smooth cp Ti screws, which had a low BIC during the healing time, caught up with the initially higher BIC of rough-surface screws during functional loading.

This speculation could be applied to the interpretation about bone area, too. Bone area of Mg-implants was not significantly different with control groups. Ellingsen's experiment in rabbit tibiae model showed similar assumption to this study.[24] Evaluating the effect of fluoride modification, he found no significant difference in percentage of bone area in the threads at 1 month, but after 3 months the TiO_2 surface had a significantly higher percentage of bone in the threads than fluoride modified TiO_2 surfaces. He described the possible cause as follows; since the control implants had less bone-to implant contact, the increased bone mass at a distance from the TiO_2 surface may be explained as a biomechanically regulated compensation in bone support.

Overall, the previous studies reported that bioactive surface chemistry of Mg might favor fast and strong integration of implants in bone in an early healing period. However, the present study showed that the osseointegration of Mg-implants under delayed loading resembled to that of turned implants. When synthesizing these results, we might suppose that Mg-implants may enhance the possibility of immediate or early loading and provide bone responses similar to biocompatible cp Ti implants in the long term. However, further investigations is needed to reveal the bone responses to Mg-implant in any other species under diverse loading period. Still more, it is considered that further clinical studies on Mg-implant will be needed.

CONCLUSION

The histomorphometric and stability analysis of Mg-implants and turned implants were done in the beagle dogs after delayed loading. In the limitation of this study, bone-to-implant contact (BIC) and bone area of Mg-incorporated oxidized implant were similar to machine-turned implant. The stability analysis showed no significantly different ISQ values and marginal bone loss between two groups. Considering time-dependent bone responses of Mg-implant, it seems that Mg-implants enhanced bone responses in early loading conditions and osseointegrated similarly to cp Ti implants in delayed loading conditions. However, further investigations are necessary to obtain long-term bone response of the Mg-implant in human.

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하중을 가한 두 가지 표면의 임플란트에 관한 조직형태학적 분석 및 안정성 분석 (비글견을 이용한 연구)

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최적의 골유착을 얻기 위하여 임플란트 디자인의 개선과 다양한 표면처리 방법이 개발되어 왔는데, 특히 최근 알칼리 에칭과 이온 주입법, 양극산화법 등 생화학적인 골유착을 유도할 수 있는 표면개질 방법이 관심의 대상이 되고 있다. 이러한 방법 중 마그네슘 이온을 함유한 전해액속에서 양극산화피막처리한 임플란트(마그네슘 임플란트)를 이용하여 조기 하중의 가능성이 제시된 바 있다. 그러나 마그네슘 임플란트의 경우 장기간의 기능적 하중을 가한 경우에 대한 연구가 부족한 상태이므로, 이에 본 연구에서는 비글견을 이용한 동물실험을 통해 지연하중을 가한 마그네슘 임플란트의 방사선적, 임상 안정성 검사 및 조직형태학적 분석을 시행하여 화학적 표면개질 임플란트의 골조직 반응을 기계가공 임플란트와 비교 평가하고자 하였다.

발치 후 3개월의 치유과정을 가진 비글 성견 6마리의 하악에 좌우 3개씩 실험용으로 제작된 직경 3.75 mm, 길이 10.0 mm의 나사 형태 임플란트 36개를 보편적인 임플란트 시술시 이용하는 식립법을 이용하여 식립하였다. 18개의 대조군은 기계가공만 한 상태였고, 실험군은 마그네슘을 함유한 전해액에서 양극산화 피막처리하였다. 식립 후 3개월 동안의 치유기간 후 이차수술을 실시하였으며, 연결고정하지 않고 단일치 금관수복으로 보철물을 장착하여 하중을 가하였다. 3개월 동안 하중을 가한 후에 희생시켜 비탈회 연마 표본을 제작하여 조직형태 계측학적 분석을 시행하였다. 임플란트 식립시, 이차 수술시, 하중을 가한 1개월, 3개월 후에 변연골 흡수를 평가하기 위해 방사선학적 검사를 실시하였고, 골계면 사이에서의 안정성을 평가하기 위해 공진주파수 수치를 측정하였다. Mann-Whitney U test와 repeated measured ANOVA를 이용하여 95 퍼센트 유의수준으로 통계적 유의성을 확인하였다.

총 36개의 임플란트 중 8개에서 일차 수술 후 골유착의 실패가 나타났으며, 1개는 3개월의 부하 후에 실패 양상이 나타났다. 공진주파수분석결과, 마그네슘 임플란트군은 대조군과는 달리 공진주파수 수치가 증가하다가 다소 감소하는 양상을 보였으며, 하중을 가한 3개월 후에는 두 군의 수치가 비슷하였다. 방사선 사진 분석 결과 두 군 모두 시간에 따른 변연골 흡수량이 증가하였으며, 통계적으로 유의한 차이는 없었다. 조직형태학적인 분석 결과 마그네슘-임플란트가 대조군에 비해 더 높은 수치의 골임플란트 계면접촉율을 보여 주었으나, 나사산내 골면적은 더 낮았다. 그러나 두 군 모두에서 통계적 유의성은 없었다.

이상의 결과에서 지연 하중을 가하는 경우에 있어서 골유착에 대한 마그네슘-임플란트의 효과는 기계가공된 임플란트와 유사하였다. 조기 하중시 더 빠르고 강한 골반응을 보여주던 이전 연구와 종합하여 볼 때, 마그네슘 임플란트는 즉시 또는 조기 하중 가능성을 증진시켜 주며, 지연 하중에서는 생체 적합성이 우수한 타이타늄과 유사한 골유착 정도를 보이는 것으로 사료된다. 그러나 임상적으로 화학적 표면개질 방법의 유용성을 판단하기 위해 다양하고 장기적인 임상 연구가 필요하리라 사료된다.

주요어: 하중, 생화학적 결합, 마그네슘-임플란트, 공진주파수 분석, 골임플란트계면 접촉율, 골면적

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원고접수일: 2008년 3월 18일, 원고수정일: 2008년 12월 03일, 원고채택일: 2008년 12월 25일