Biological Evaluation of Implant Drill Made from Zirconium Dioxide

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ABSTRACT

Purpose: Zirconia is a good candidate material in the dental field. In this study, we evaluated biological responses against a zirconia drill using a bone cavity healing model.

Materials and Methods: Zirconia drills, stainless steel drills, and the drilled bone surface were observed by scanning electron microscopy (SEM), before and after cavity preparation. For the bone cavity healing model, the upper first and second molars of Wistar rats were extracted. After 4 weeks, cavities were prepared with zirconia drills on the left side. As a control, a stainless steel drill was used on the right side. At 3, 7, and 14 days after surgery, micro-CT images were taken. Samples were prepared for histological staining.

Results: SEM images revealed that zirconia drills maintained sharpness even after 30 drilling procedures. The bone surface was smoother with the zirconia drill. Micro-CT images showed faster and earlier bone healing in the zirconia drill cavity. On H-E staining, at 7 days, the zirconia drill defect had a smaller blank lacunae area. At 14 days, the zirconia drill defect was filled with newly formed bone.

Conclusions: The zirconia drill induces less damage during cavity preparation and is advantageous for bone healing. (197 words)

KEY WORDS: bone defect healing, dental implant, osseointegration, zirconium dioxide

INTRODUCTION

Dental implants represent an important prosthetic treatment option and the success rate of dental implant treatment is relatively high. Nonetheless, early implant failures, i.e., osseointegration failures of unknown cause, continue to occur, and it is important to minimize this early failure rate.

Quality and quantity of bone surrounding implants and primary stability of implants are important elements for the establishment of osseointegration, which is essential for implant success. At the same time, inflammation control and acceleration of bone healing after cavity preparation play a pivotal role in implant therapy. In this regard, minimal bone damage upon cavity preparation would help to improve success rate, and it has been noted that external irrigation during implant cavity formation results in less heat and damage to surrounding bone.

Zirconium dioxide, which is known as zirconia, is a good candidate material in the dental field, because of its physical properties and biocompatibility. Zirconia has mainly been used for implant abutments and superstructures because of its durability, strength, and corrosion resistance. At the same time, several implant drills are already commercially available for the same reason. Ceramic composite drills containing zirconium oxide were able to maintain their drilling ability and maintain lower temperatures than stainless steel drills.
drills during cavity preparation in an in vitro study. More recently, zirconia has also been used as a fixture material because of its biocompatibility and physical properties. Moreover, zirconia has been reported to be a very good material for biopsy needles, as it induces less inflammation than stainless steel needles. Thus, zirconia might be a good material for implant drills. However, detailed comparisons of biological responses, such as bone healing after drilling, between drills made from zirconia and other materials have not been reported. In this study, we evaluated the biological responses against zirconia implant drills using a rat bone defect healing model and compared it with that of stainless steel implant drills.

**MATERIALS AND METHODS**

**Implant Drills**

Custom-made zirconia and stainless steel implant drills specifically designed for rats were prepared (Pilot Corporation, Japan). Both drills were identical in diameter (1.7 mm), drill point, blade, and shape. They were used under exactly the same conditions. SUS402J2 stainless steel, as stipulated by Japanese Industrial Standards, was used for the stainless steel drill. SUS402J2 contains less than 0.26 ~0.4% carbon, less than 1% silicon, less than 1% manganese, less than 0.04% phosphorus, 0.15% sulfur, 12~14% chromium, and 84~86% iron.

**Scanning Electron Microscopy (SEM) Analysis**

In order to investigate the sharpness of the drills and texture of the surface of drilled bone after cavity preparation, SEM analysis was carried out. A Philips XL20 (Philips, Eindhoven, Netherlands) electron microscope was used for image analysis. Before drilling and after drills were used 10 or 30 times for cavity preparation in rat femoral bone, the major cutting edge, cutting edge corner, and minor cutting edge of the drills were analyzed (Figure 1). The major cutting edge, cutting edge corner, and minor cutting edge of the drills contribute to drilling, while the chisel edge does not have cutting capacity.

**Cavity Healing Model**

Thirty-six 4-week-old male Wistar rats (Charles River, Yokohama, Japan) were used for cavity healing model. For preparation of the cavity healing model, upper first and second molars on both sides were extracted under anesthesia by intraperitoneal injection of 8% chloral hydrate (400 mg/kg). Four weeks after tooth extraction, bone cavities were prepared at the first molar site. Initially, a small pit was made using a round bur as a center punch for the main drill. Main bone cavities were prepared with a custom-made zirconia drill on the left side of rat maxilla at 800 rpm with external irrigation. Each drill was used less than 10 times. As a control, defects were prepared using a similarly shaped custom-made stainless steel drill on the right side in the same animal. Flaps were repositioned and sutured with nylon thread (Figure 2). All animal experiments in this study belonged to category C in the SCAW, were approved by the Ethics Committee of Niigata University, and were conducted in accordance with the Niigata University Guidelines for Animal Experimentation.

**Micro-Computed Tomography (Micro-CT) Analysis**

At 7 and 14 days after cavity preparation, rats were sacrificed and perfused with 4% paraformaldehyde. Maxillary bone samples were collected and analyzed by micro-CT scanner (Elescan, Tokyo, Japan) to observe newly formed bone. The region of interest (ROI) was the area of the bone cavity which was made at first molar area. Briefly, the maxillary bone was placed on a custom-made jig with the axial direction and palatal area facing toward the scanner. Scanning was performed at 53 kV, 100 lA, and 900 projections, with a 0.5-mm aluminum filter. Based on the serial-scanned images, 3D images were reconstructed using TRI/3D-BON software (RATOC, Tokyo, Japan).
Histological Investigation

Animals were sacrificed at 3, 7, and 14 days after cavity formation. At the appointed times, they were anesthetized and fixed with a transcardiac perfusion with a fixative containing 4% paraformaldehyde (pH 7.4). Specimens were decalcified in 10% EDTA solution for 4 weeks at 4°C. Serial paraffin sections were prepared sagittally and horizontally at 5 μm thickness, and

Figure 2 Time course of experimental procedure.

Figure 3 Scanning electron microscope image of stainless drill point (A, C, E) and zirconia steel drill point (B, D, F).
sections from the most central part of the defect were selected and stained with hematoxylin and eosin (H-E) for histological observation and for histochemistry with tartrate-resistant acid phosphatase (TRAP) for osteoclast staining. Each time point had 12 rats. For sagittal sections, six rats were used and five sections were selected from the central area of the cavities. For horizontal sections, six rats were used and five sections were selected from each cavity. From 30 sections in each time point, number of blank lacunas, number of TRAP positive cells and areas of newly formed bone were measured using MetaMorph software (Universal Imaging Corporation, West Chester, PA).

Statistics
Data are expressed as means and standard deviation (SD). Student’s t-test was performed to analyze the differences between two groups. *p* Values of less than 0.05 were considered to be significant.

RESULTS
SEM Images of Drills and Drilled Bone Surface
Before drilling (0 times), both the zirconia drill and stainless steel drill had a sharp major cutting edge and cutting edge corner, and they looked identical (Figure 3, A and B). After 10 drilling procedures in 12-week-old rat femoral bone, SEM images showed no wear or deformation on either the zirconia or stainless steel drill at the chisel edge, major cutting edge, or cutting edge corner (Figure 3, C and D). However, after 30 drilling procedures, although the zirconia drill kept its sharpness (Figure 3F), the stainless steel drill showed some blunting and small notched ends on the major cutting edge and cutting edge corner (Figure 3E). In terms of the minor cutting edge, similar to the major cutting edge and cutting edge corner, the stainless steel drill showed durability for 10 drilling procedures (Figure 4, A, C, and E), but the zirconia drill maintained its sharpness.

![Figure 4](image-url)

**Figure 4** Scanning electron microscope image of minor cutting edge in stainless drill (A, C, E) and zirconia drill (B, D, F).
for 30 drilling procedures (Figure 4, B, D, and F). Based on these findings, drills were used for less than 10 procedures in subsequent in vivo experiments and observations of drilled femoral bone surface. Regarding the drilled bone surface structure analyzed by SEM, stainless steel drilled bone under low magnification showed an irregular cavity edge (Figure 5A). In contrast, the zirconia drilled bone showed a clear cut edge (Figure 5B). Under high magnification, the stainless steel drill defect showed a rough and microscaled surface (Figure 5C). On the other hand, zirconia-drilled bone showed a smooth and flat surface (Figure 5D).

**Micro-CT Image Analysis of Cavity Healing**

Micro-CT images of the maxillary bone 7 days after defect formation showed that there was no new bone formation from the cavity edge and no differences between stainless steel (Figure 6A) and zirconia-drilled bone cavities (Figure 6B). At 14 days after cavity preparation, new bone formation from the edge of the defect could be seen in both samples. However, the amount of newly formed bone was larger, and faster defect healing was observed, in the zirconia drill defect (Figure 6D) than in the stainless steel drill defect (Figure 6C). The zirconia drill cavity showed significantly higher new bone formation capacity than stainless steel drill cavity on day 14 (stainless steel drill, $2.111 \pm 0.218$ versus zirconia drill, $2.506 \pm 0.168$ $p < 0.05$).

**Histological Analysis of Drilled Bone**

There were no differences in bone formation between the stainless steel drilled cavity (Figure 7A) and zirconia-drilled cavity (Figure 7B) at 3 days after cavity preparation. Seven days after drilling, low magnification images showed no significant differences (Figure 7, C and D), but high magnification images confirmed that the zirconia drill cavity (Figure 7, F and G) had a thinner blank lacunae area (double-headed arrow) than the stainless steel drill cavity (Figure 7, E and G) (stainless steel drill, $179.166 \pm 28.637$ vs zirconia drill, $101.777 \pm 12.558$ $p < 0.05$). At 14 days after cavity preparation, even though new bone formation could be seen in both cavities (Figure 8, A and B), more new bone formation was observed in the zirconia drill cavity (Figure 8D) than stainless steel drill cavity (Figure 8C) on horizontal images. Histometric analysis showed that significant increase of newly formed bone in zirconia drill cavity compare to stainless steel drill cavity.
(Figure 8G) (stainless steel drill, 2,034 ± 0.323 versus zirconia drill, 3.588 ± 0.410 \( p < 0.05 \)).

In TRAP staining images at 14 days after cavity preparation, more TRAP-positive cells were observed at the boundary between newly formed bone and existing bone in the stainless steel drill cavity (Figure 8, E and H) than in the zirconia drill cavity (Figure 8, F and H) (stainless steel drill, 16.308 ± 3.923 versus zirconia drill, 4.538 ± 1.330 \( p < 0.05 \)).

**DISCUSSION**

Harder and colleagues showed that the method of cooling affects the development of intrabony temperature during preparation of the implant site, but the drill material appears to play no particular role. Oliveria and colleagues. reported that both the stainless steel drill and the zirconia drill could be used up to 50 times without producing harmful temperatures in the bone tissue or severe signs of wear and deformation. However, our SEM image analysis showed that even though both drills had no substantial wear or deformation even after use, drilled bone surface structures revealed different features between zirconia and stainless steel after fewer than 10 procedures.

In this study, we established a bone cavity healing model using rat maxillary bone. Several
studies have noted that the heterogeneity of osteoblasts and osteoclasts depends on bone site. The reason that we selected maxillary bone for the animal model was that we would apply these findings to clinical oral implant therapy as a final aim of our study.

Figure 7 H-E staining image of rat maxilla at 3 and 7 days after cavity preparation. (A, C, E) were prepared with the stainless steel drill and (B, D, F) were prepared with the zirconia drill. (A–D) are low magnification (2.5×) and (E, F) are high magnification (40×). Blank lacunae spaces were measured and compared in (G).
Osteocytes that possess a lacuna-canalicular network in the bone substance are thought to contact one another and are very sensitive to stimulation. For example, a mere periosteum reflection results in osteocyte cell death through the lacuna-canalicular network, which may be observed as blank lacunae.\(^{10}\) In our

![Figure 8](image-url)
histological images, a thicker blank lacunae area was observed with the stainless steel cavity than the zirconia cavity. This suggests that preparation with the stainless steel drill had a greater impact on osteocytes, even without heat or other stimulation. Because the surface of cut bone with the stainless steel drill was rougher, stimulation may affect osteocyte cell death and inhibit bone formation on the surface thereafter. Moreover, larger numbers of osteoclasts were observed after drilling with the stainless steel drill than with the zirconia drill. Even though this finding may not directly affect bone defect healing, it is clear that there was a difference in cell and tissue responses between these two drills regardless of drill sharpness or wear.

Previous zirconia implant studies confirmed its bone tissue conductive abilities, which is almost equivalent to those of titanium implants.\(^{11–13}\) In the case of soft tissue, zirconia needles are able to accelerate stem cell recruitment when compared to stainless steel needles.\(^6\) Stem cell recruitment is an important factor for new bone formation.\(^{14}\) We were unable to clearly identify the possibility of unknown zirconia effects with the present results. However, if zirconia drills also possessed stem cell recruitment ability, they might become more reliable drilling instruments for implant therapy. To confirm this hypothesis, comparative single drill use experiments for each material are currently in progress.

A mechanical engineering study and surgical instrument development study showed that the cutting efficiency is influenced by design, shape, drill materials, and drilled subjects.\(^{15,16}\) Therefore, the most effective designs for zirconia and stainless steel drills may be different. Our results indicate the superiority of zirconia drills for bone healing period, even if their design is not ideal for drilling efficiency. As the drilling efficiency may be closely related to the establishment of osseointegration,\(^{17,18}\) the improvement of zirconia drill shape would benefit the clinical outcome of implant therapy. Further study into the optimal shape and design for zirconia drills remains necessary. In conclusion, zirconia drills were able to induce bone healing after implant cavity preparation more effectively than stainless steel drills. This study suggests that zirconia drills may help in the establishment of osseointegration, which would reduce early implant failures.

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