

Effect of Implant Site Preparation by Piezoelectric and Conventional Drilling on Autograft Cell Viability: A Clinical Trial

J. Moradi Haghgoo¹, S. Soheilifar¹, M. Nikkhah², P. Torkzaban³, N. Rabienejad¹ 

¹ Assistant Professor, Department of Periodontics, School of Dentistry, Hamadan University of Medical Sciences, Hamadan, Iran

² Assistant Professor, Department of Periodontics, School of Dentistry, Guilan University of Medical Sciences, Guilan, Iran

³ Associate Professor, Department of Periodontics, School of Dentistry, Hamadan University of Medical Sciences, Hamadan, Iran

Abstract

Background and Aim: Implant site preparation with minimal trauma is an important factor in success of implant treatment and has a great effect on osseointegration. Piezoelectric bone surgery was introduced as a modern technique for micrometric osteotomy with precise, controllable action. Preservation of osteoblast cell viability is critical to achieve successful osseointegration. The present study was carried out to evaluate the effects of two implant site preparation methods on cell viability of bone particles collected during osteotomy.


Materials and Methods: In this clinical trial, 45 samples of bone chips were collected during implant site preparation by conventional drilling and 45 samples were collected by piezosurgery. Cell viability of bone chips collected by osteotomy was evaluated using MTS kit in both groups. Data were analyzed by t-test and Kolmogorov-Smirnov test via SPSS version 21 software.

Results: Findings showed that the percentage of cell viability in the piezosurgery group (54.40%±7.71%) was greater than that in the conventional drilling group (29.93%±6.08%) and this difference was statistically significant (P=0.000).

Conclusion: Findings of the present study show that bone particulates collected by piezoelectric system have greater potential for longevity than those collected by conventional rotary system and can enhance bone healing around implants and result in successful osteointegration.

Key Words: Dental Implants, Cell Survival, Osteogenesis, Osseointegration, Piezosurgery

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 Corresponding author:
N. Rabienejad, Assistant Professor, Department of Periodontics, School of Dentistry, Hamadan University of Medical Sciences, Hamadan, Iran

nazlirabi@yahoo.com

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Introduction

In the recent years, patients' demand for dental implant treatment has greatly increased due to high satisfaction rate of patients with this modality. Dental implants as a replacement for the missing teeth provide optimal esthetics, speech, chewing function and self-confidence and improve the quality of life [1,2].

Creation and continuation of osseointegration is fundamental for dental implant success. Many factors affect osseointegration such as the implant-bone contact, biocompatibility, implant surface properties and surgical technique [3]. Accurate surgical technique and proper implant site are among the important clinician-related factors. Attempts have been made to decrease

trauma during implant site preparation especially heat trauma.

Piezosurgery uses an ultrasonic transducer. This modality has implications in maxillofacial, cerebrospinal, hand and foot surgeries [4,5]. Low surgical trauma, better surgical control and fast tissue healing are some of the benefits of piezosurgery. Minimal surgical trauma induces tissue regeneration mechanisms while high surgical trauma can result in scar tissue formation [6,7].

From the mechanical point of view, spiral drills or burs cause lamellar fracture on surfaces beside the cutting edges. Large bone particles deposit at endosteal spaces and lead to delayed osteogenesis. Micromechanical cutting in piezosurgery causes micronization of bone particles and does not cause lamellar fracture. Piezosurgery results in release of bone morphogenetic protein, which is an affective bone regeneration factor. Decreased tissue inflammatory response is also considerable with piezosurgery [7,8].

The quality of implant site preparation and its effect on treatment results have not been well evaluated in previous studies [9-13]. The present study was carried out to evaluate the effects of two implant site preparation methods on cell viability of bone chips collected during osteotomy.

Materials and Methods

In this clinical trial, 45 patients (17 males and 28 females, age range of 30-55 years) referred to the Periodontics Department of Hamadan University, Dentistry Faculty requiring dental implant treatment were evaluated. They were informed about all the procedures, and written informed consent was obtained from them. This study was approved by the ethics committee of Hamadan University of Medical Sciences (IR.UMSHA.REC.1394.248) and registered in the Iranian Registry of Clinical Trials (IRCTID: IRCT2015102624726N1).

The inclusion criteria were as follows: good oral hygiene, two implant sites next to each other, and at least six months had passed since the tooth extraction.

The exclusion criteria were as follows: systemic diseases, pregnancy, smoking, alcohol consumption, periodontal disease, radiotherapy,

chemotherapy, taking immunosuppressive drugs and history of bone regeneration at the implant sites.

The test and control specimens were collected from each patient with D2 and D3 bone density according to patients' cone beam computed tomography scans [14]. Allocation of groups to the sites was done randomly by flipping a coin.

The surgical procedure was done as explained below:

The patients rinsed 0.2% chlorhexidine gluconate mouthwash for one minute to decrease the bacterial count. A crestal incision was made after anesthesia induction by injecting 2% lidocaine solution containing 1:100,000 epinephrine.

The control specimens were obtained by implant surgery micromotor (Surgic XT Plus; NSK, Tokyo, Japan) using the first drill at 800 rpm speed. Irrigation was done by saline, and the drill was used with pumping motion to minimize heat generation. Bone particles were collected by Kohler Bone Aspirator (Kohler, Neuhausen, Germany). We used the same first drill for bone harvesting in the control group.

The test specimens were obtained by piezosurgery (Variosurg, NSK, Tokyo, Japan) at 35 kHz frequency. Bone harvesting was done by SG15A head (first drill). Osteotomy was performed with gentle pressure under saline irrigation. Bone particles were collected by Kohler Bone Aspirator (Kohler, Neuhausen, Germany). The test group specimens were all harvested with the same settings of the piezosurgery device.

Bone chips were collected by the aspirator mounted on the surgical suction to eliminate saliva contamination. We used another surgical suction to collect patient's saliva. All surgical procedures and sample collection were done by a postgraduate student of periodontics. The acquired samples were kept in microtubes containing 0.9% saline. The samples were placed on ice and immediately sent to a laboratory.

MTS kit was used to evaluate cell viability [3-(4,5-Dimethylthiazol-2-yl)-5-(3-carboxymethoxyphenyl)-2-(4-sulfophenyl)-2H-tetrazolium, inner salt or MTS reagent; ABCAM, Houston, USA]. MTS kit can accurately determine osteoblast proliferation and cell viability. Approximately, 100 mg of autogenous bone

samples was used according to a study by Miron et al [15]. The bone samples were irrigated by phosphate buffered saline 3 times to eliminate other cells such as blood cells. Then, the bone chips were converted to fine particles by a bone mill. The samples (containing osteoblastic cells, 80 μ L of MTS solution and 400 mL phosphate buffered solution) were incubated at 37°C for 4 hours. Viable cells produce formazan crystals by mitochondrial function [NADP(H)-dependent dehydrogenase]. After 4 hours, the microtubes were placed in a spectrophotometer to evaluate cell viability. The absorbance of colored solution was measured at 490 nm wavelength and the optical density was reported [15]. Data were analyzed using t-test and Kolmogorov-Smirnov test via SPSS version 21 (SPSS Inc., IL, USA).

Results

In the present study, cell viability was compared in bone particles collected after conventional drilling and piezosurgery during implant site preparation. A total of 45 autogenous bone samples were obtained in each group (control and test). Kolmogorov-Smirnov test showed normal distribution of data. Thus, t-test was used for data analysis.

The mean optical density of the colored solution in piezosurgery group was higher than that of the drilling group (Table 1, Diagram 1).

As shown in Diagram 1, the minimum cell viability was 12% in the conventional drilling group and 34% in the piezosurgery group. The maximum cell viability was 43.7% in the conventional drilling group and 66.5% in the piezosurgery group. The t-test showed a significant difference in the mean cell viability between the two groups ($P=0.000$).

Discussion

Osseointegration is one of the most important clinical goals of implant treatment. Successful osseointegration depends on osteoblastic cell viability at the surgical site and osteogenic differentiation and proliferation of mesenchymal cells. Piezosurgery causes minimal hard and soft tissue damage by selective precise cutting [4-6,8,16]. Most previous studies have been conducted on animals with histological tissue evaluation. Thus, we evaluated the implant site

preparation techniques and bone response in humans.

Our study showed preservation of cell viability in bone particles of both site preparation approaches using MTS kit. Although Gruber et al, [17] in 2005, Kuttenger et al, [18] in 2013, Chiriac et al, [19] in 2005 and Miron et al, [15] in 2013 used different techniques but found the same result.

Cell viability was significantly greater in the piezosurgery group in our study. Berengo et al. [20] reported superior results for bone harvesting by piezosurgery because of absence of non-vital bone; the same findings were also reported by Von See et al, [21] in 2010 and Hsu et al, [22] in 2011. But, Berengo et al. [20] evaluated the samples using histological analysis. Von See et al. [21] used animal samples and evaluated them under a microscope. Only Hsu et al. [22] used cell viability kit for animal samples.

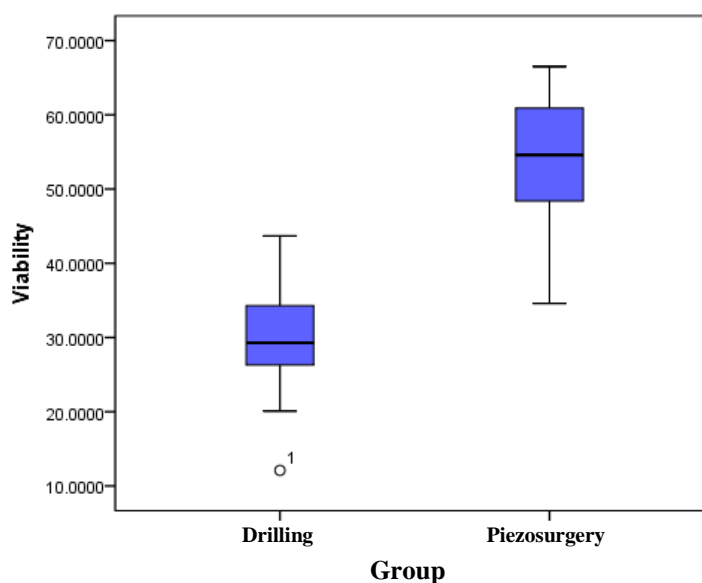
Better cell viability in piezosurgery group may be related to vibration movements that are converted to energy and less pressure of hand piece causing insignificant thermal alteration. Micromechanical cutting results in release of bone morphogenetic proteins [4-6]. Piezosurgery osteotomy surface has a narrower necrotic layer [23]. All these factors contribute to less surgical trauma and better tissue healing.

Despite all the above, some studies reported equal osteogenic capability of piezosurgery and drilling [15, 24, 25]. The samples were collected from the cortical bone in the study by Chiriac et al, [19] which causes over heating (degradation of proteins and lipids, alteration of enzymes). We collected D2 and D3 bone samples to have less thermal alteration and obtain more viable cells.

A proper technique should be used to evaluate cell viability. Histologic and histomorphometric evaluations are not suitable for this purpose. Miron et al. [15] used the same method as ours but reported different results. This difference may be due to variability in bone density, inappropriate cutting pressure and inadequate irrigation. The quality (sharpness) of bur can also affect the results. In this study, we used new sharp burs to have less thermal alteration [26]. The quality of transfer of samples and collection of saliva is another important factor that has not been mentioned in other studies. We used a separate

Table 1. Mean optical density (percentage of cell viability) of the groups

Group	Number	Average(%)	Standard Deviation
Conventional drilling	45	29.93	6.08
Piezosurgery	45	54.40	7.71
Total	90	42.16	14.11

**Diagram 1.** Mean optical density (percentage of cell viability) of the two groups

surgical suction and chlorhexidine mouthwash to reduce bacterial load. Patients with periodontal disease were excluded from the study.

Piezosurgery also has positive effects on blood circulation in the tissue, cell migration and type I collagen formation [22]; thus, it may lead to proper tissue healing after flap surgery and tissue regeneration.

As reported by Esposito et al, [27] in their study on immediately loaded zygomatic oncology implants in edentulous maxillae, both conventional and piezosurgery drilling yield the same clinical results; whereas, conventional drilling is more aggressive and needs less time. It is noteworthy that the outcomes are probably system-dependent

and therefore, the results cannot be generalized to all conditions [27].

The strengths of our study were large sample size, human osteoblast samples and using a precise cell viability kit.

Conclusion

The results of this study showed higher cell viability in piezosurgery group. These active cells enable better healing and osseointegration. Thus, better treatment outcomes may be expected by using piezosurgery. Considering better cell viability in piezosurgery group, this technique can be used for regeneration of bone defects.

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