



Evaluation of the efficacy of using dental pulp graft in the healing of the alveolar bone after impacted canine extraction: a prospective cohort study

Omar Aljarmakani, MSD*, Mounzer Assad, PhD

Summary: The purpose of this study was to evaluate the radiographic density of the alveolar bone of the maxilla after extraction of the impacted canines and using the pulp tissue as an autogenous graft.

Materials and methods: This prospective cohort study recruited 14 patients (8 females and 6 males) between 2021 and 2023, with an average age of 35 years. All participants had palatally impacted maxillary canines. The impacted teeth were extracted surgically. The extracted teeth were then used for autogenous grafting. The pulp tissue was removed, cut into small pieces, and placed on an absorbable gelatin sponge before being inserted into the extraction socket. The wound was subsequently closed meticulously. After 4 months, the bone density was assessed radiographically using the Hounsfield Scale on cone beam computed tomography scans.

Results: After 4 months, the mean radiographic bone density value in the extraction area was (652.77 ± 56.13 HU), while the average density of the original bone was (659.7 ± 39.6 HU).

Conclusions: Within the limits of this study, dental pulp tissue can be used to restore bony defects of the alveolar bone in the maxilla. However, further research is needed to confirm these findings.

Keywords: stem cells, bone grafts, dental pulp

Introduction

Background and objectives

The continuous increase in tissue loss due to various illnesses and injuries significantly impacts the quality of life for individuals across different societies^[1]. Restoring bony defects and rebuilding bone remains a major clinical challenge, particularly in dentistry. Tooth loss, periodontal disease, trauma, tumour resection, congenital defects, and actinic osteonecrosis are leading causes of bone loss in the face and jaws. Periodontal disease and bone remodelling following tooth loss are the most common causes of alveolar bone defects^[2,3].

Bone resorption occurs after teeth extraction, resulting in a loss of alveolar bone volume and a change in the shape of the alveolar ridge, which can affect subsequent implant placement^[4,5]. In order to maintain adequate levels of bone and soft tissues for

HIGHLIGHTS

- This technique provide a new and effective intervention for repair alveolar bone defects by using the dental pulp tissue as material graft.
- This technique is a predictable and effective regenerative procedure for patients with acquired alveolar bone defects to obtain a new bone with a good density in a good time.
- This technique is easy and did not need any laboratory works.

aesthetic purposes and long-term success, several techniques have been developed to reconstruct hard and soft tissues^[6,7].

In order to minimize the need for guided bone regeneration after teeth extraction, techniques for socket preservation have been developed. These techniques aim to reduce the loss of hard and soft tissues during healing by using bone substitutes in the extraction socket. Several techniques and materials have been used to maintain the size of the alveolar bone after extraction^[8,9]. Although several biomaterials and growth factors have been used, there is currently no consensus on the ideal technique and materials^[10,11].

The search for better bony alternatives that possess the three key characteristics of the ideal bone graft (osteoconduction, osteoinduction, and osteogenesis) is still continues. While autografts exhibit all these properties, the need for a second surgical site, associated pathology, and limited available material have driven researchers to explore alternative sources like xenografts and alloplasts^[12,13]. However, xenografts carry a risk of disease transmission, and alloplasts, while readily available, are expensive and often deliver unsatisfactory results^[14]. Their lack of osteoinduction limits their use to filler materials^[15,16].

Department of Oral and Maxillofacial Surgery, Faculty of Dentistry, Tishreen University, Lattakia, Syria

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*Corresponding author. Address: Department of Oral and Maxillofacial Surgery, Faculty of Dentistry, Tishreen University, Lattakia, Syria., Tel.: +963 991 786 170. E-mail: omar.aljarmakani@tishreen.edu.sy (O. Aljarmakani).

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Growth factors are a group of cytokinase that can stimulate specific biological responses, including proliferation, differentiation, and migration of various cell types, some of these growth factors include bone morphogenetic protein (BMP), platelet-derived growth factor (PDGF), and vascular endothelial growth factor (VEGF)^[17,18]. These growth factors have been used in conjunction with bone grafts to preserve the alveolar ridge after teeth extraction^[19]. The growth factor BMP has a high ability to stimulate bone formation, as it can stimulate undifferentiated mesenchymal cells to differentiate into osteoblasts. Similarly, the growth factor PDGF stimulates both mesenchymal cells and osteoblasts, although its ability to form bone is less than that of BMP^[20].

Sasaki and Watanabe have observed that hyaluronic acid (HA) is capable of accelerating the formation of new bone through the differentiation of mesenchymal cells, and they have demonstrated stimulation of bone formation on the four day after HA application. HA possesses biochemical and physical properties that play an important role in the early stages of bone formation^[21].

A new concept has emerged that encourages bone rebuilding using tissue engineering, offering an alternative solution to traditional bone grafting. This approach utilizes living components capable of integrating with surrounding native tissue^[22]. It relies on isolating cells from the tissue and expanding them *in vitro* through bioreactors and three-dimensional (3D) scaffolds^[23]. The use of dental pulp stem cells in rebuilding bony defects is considered one of the latest tissue engineering techniques, holding promise for revolutionizing future treatment modalities^[24].

Despite the significant advancements in this field, the large discrepancy in research results makes reaching a definitive conclusion challenging. Additionally, the clinical application and long-term effectiveness of stem cell-based therapies are not yet fully established^[25].

Materials and methods

Ethics

This research was conducted with ethical approval number (1362) dated 25-2-2020 and in accordance with the Declaration of Helsinki for human studies. This work also adheres to the STROCSS 2021 criteria^[26]. This research was registered at Research Registry under the identifying number: researchregistry9558. https://www.researchregistry.com/browse-the-registry#home/?view_2_search=omar%20aljarmakani&view_2_page=1.

Purpose of the study

This study aimed to restore the alveolar bone after extraction of the impacted upper canine by applying dental pulp tissue as an autogenous graft in the alveolar socket. Radiographic bone density was evaluated at four months post-surgery.

Research sample

This prospective cohort study included 14 patients (8 females and 6 males) with palatally impacted upper canines diagnosed by occlusal radiography and deemed unsuitable for orthodontic traction. The study was conducted between 2021 and 2023. Participants were between 31 and 40 years old, with an average age of 35 years. Medically healthy patients were selected based on a completed questionnaire and written informed consent. All

patients received follow-up radiographic imaging [cone beam computed tomography (CBCT)] 4 months after surgery. Patients with a history of heavy smoking or compromised systemic diseases were excluded from the study.

The patients were evaluated seven days before surgery. They received a gingival scaling and were instructed on how to perform proper oral hygiene, including the use of chlorhexidine 0.12% mouthwash twice daily after brushing until the day of surgery.

Extraction of the impacted canine

All surgical procedures were performed by the same surgeon. Prior to the procedure, the patient rinsed their mouth with chlorhexidine 0.2% for one minute. Local anaesthesia was administered using lidocaine 2% with epinephrine 1:80 000 as a vasoconstrictor. In cases requiring general anaesthesia, it was administered accordingly. A full-thickness palatal envelope flap was raised using a surgical blade #15 by making a horizontal incision around the cervix of the teeth. The flap was dissected at the subperiosteal level with a periosteal elevator (Malt9), taking care not to damage the nasopalatine neurovascular bundle (Fig. 1).

The impacted canine tooth was exposed by creating a bony window in the hard palate using a rounded surgical bur (tungsten carbide) on a straight handpiece under continuous irrigation with saline solution 0.9% (Fig. 2). The canine was then fully extracted using a straight elevator (Fig. 3).

The bone edges were smoothed with a bone file. The extracted tooth was immersed in chlorhexidine 2% for one minute for disinfection. The crown was then separated from the root by creating a groove at the cemento-enamel junction using a flat-end diamond bur on a turbine handpiece with continuous irrigation with saline solution 0.9%, ensuring no access to the pulp chamber. The crown was subsequently separated from the root using forceps. The dental pulp was harvested with a dentin excavator and barbed broaches and placed in a 2 ml vial containing saline solution 0.9% (Fig. 4).

The harvested pulp tissue was cut into pieces smaller than 0.5 mm using a surgical scalpel (Fig. 5). This mixture was then loaded onto a piece of absorbable gelatin sponge and placed into the alveolar socket of the extracted canine (Fig. 6). Following successful haemostasis, the flap was replaced and meticulously sutured using the interrupted suture technique with silk thread 0-3 (Fig. 7).



Figure 1. A clinical picture showing the flap reflection.

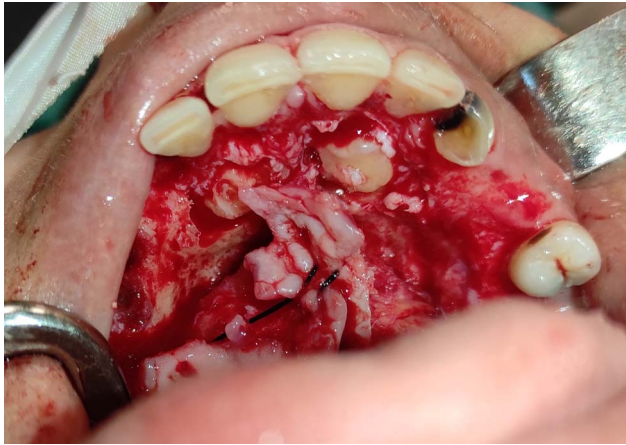


Figure 2. A clinical picture showing the impacted canine exposing.

The patient received post-extraction instructions and a drug prescription: Amoxicillin 825 mg with clavulanic acid 125 mg (Augmentin 1000 mg) for 7 days, Ibuprofen (profen 400 mg) for 3 days, and Chlorhexidine rinses 0.12% (Biofresh) for 7 days.

Evaluation after surgery

The first follow-up appointment occurred seven days after the surgical procedure to remove the sutures. The second follow-up appointment took place 4 months later, at which time a CBCT scan was performed to assess bone density. CBCT scan images were analyzed using 3D imaging software (Fig. 8).

Statistical analysis

Statistical analysis was performed using IBM SPSS Statistics version 22 (IBM Corporation, Armonk). Descriptive statistics, including the mean and standard deviation, were used to assess the bone density of the sample and the original bone. An unpaired *t*-test was employed to compare the bone density in the area where the dental pulp graft was applied to the original bone. A significance level of 5% ($\alpha = 0.05$) was used.



Figure 3. A picture showing the extracted canine.



Figure 4. A picture showing the pulp tissue in 2 ml of saline.

Results

- This study included 14 patients (8 females and 6 males) with an average age of 35 years. None of the participants withdrew from the study.
- At the first follow-up appointment seven days after surgery, no patient exhibited any signs of infection or flap dehiscence. In all cases, healing progressed normally without scarring, bleeding, oedema, or other side effects. Additionally, no patients reported experiencing postoperative pain.
- At the second follow-up appointment four months after surgery, CBCT scans were obtained to evaluate bone density where the dental pulp graft was applied. The mean radiographic bone density was 652.77 Hounsfield units (HU) with a standard deviation of 56.13 HU. The lowest and highest bone density values in the sample were 547.6 HU and 698.6 HU, respectively. The mean density of the original bone was 659.7 HU with a standard deviation of 39.6 HU. The minimum and



Figure 5. A picture showing the cutting of the pulp tissue.



Figure 6. A clinical picture showing the applying of dental pulp graft with the absorbable gelatin sponge in the extraction socket.

maximum bone density values of the original bone were 570 HU and 699.7 HU, respectively (Table 1).

There was no statistically significant difference in bone density between the original bone and the new bone [Table 2].

Discussion

Numerous materials have been used for bone grafting in dentistry, including allografts (e.g. demineralized freeze-dried allografts, freeze-dried allografts, or fresh frozen allografts), xenografts (e.g. bovine bone, coral), and alloplastic grafts (e.g. bioceramics, calcium phosphate, hydroxyapatite). However, the ideal bone graft should possess three key characteristics:

- Osteoconduction: The ability to serve as a scaffold for bone formation^[27].
- Osteoinduction: The capacity to attract cells, particularly unspecialized cells and preosteoblasts, which will form new bone^[27,28].
- Osteogenesis: The ability of cells within the graft material to differentiate and form bone^[29].



Figure 7. A clinical picture showing the suture after surgery.

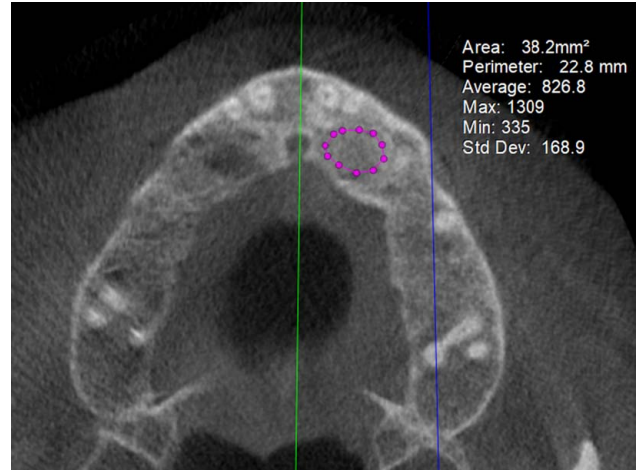


Figure 8. A radiographic cone beam computed tomography image showing the bone density of the new bone.

While allografts lack osteoinductive properties, xenografts and alloplasts possess only osteoconductive features. Autogenous grafts, exhibiting all three characteristics, are considered the gold standard^[30]. However, their use presents several challenges, including the risk of infection at the donor site, limited availability, and noticeable resorption of the graft material^[31,32].

Tissue engineering seeks to regenerate tissues by utilizing biomaterials and biomedica to develop new approaches in regenerative medicine^[33]. Despite the abundance of stem cell sources in the human body, identifying readily accessible locations to obtain sufficient quantities of stem cells remains a significant challenge in clinical applications of tissue engineering^[34–37]. While these potential sources contain an enough number of stem cells, their accessibility is limited, posing a high risk of infection and potential harm to anatomical structures. Dental pulps, containing adaptable and multi-potent stem cells derived from the neural crest, emerge as a promising donor site due to their ease of access and minimal pathogenicity^[38].

In a study by D’aquino and colleagues on 17 patients using split-mouth technique, they evaluated the effectiveness of DPSCs carried on collagen sponge in socket preservation after lower third molar extraction. They found that optimal vertical repair and complete restoration of periodontal tissues were higher in the test site compared to the control site^[33].

A systematic review conducted by Namjoynik and colleagues, which included 49 articles on the use of human DPSCs and biodegradable scaffolds in bone defect repair in animals, showed that bone regeneration was significantly higher in the experimental group (DPSCs + scaffold) compared to the control group (scaffold only). In general, there are significant differences in the response of different bone defects to human DPSCs, as skull

Table 1
Medians of bone density

| | Mean | St. deviation | Max | Min |
|---------------|--------|---------------|-------|-------|
| New bone | 652.77 | 56.13 | 698.6 | 547.6 |
| Original bone | 659.70 | 39.6 | 699.7 | 570 |

Max, maximum; Min, minimum; St., standard.

Table 2
Statistical results according to *t*-test

| <i>t</i> -test | <i>P</i> | Result |
|----------------|----------|-----------------|
| -1.04 | 0.318 | Not significant |

defects and mandibular defects showed a much higher bone regeneration response to human DPSCs compared to other defect sites^[39].

This study represents the first application of dental pulp tissue as an autogenous graft for repairing alveolar bone defects following impacted upper canine extraction in the maxilla. The obtained results provide evidence for the effectiveness of the proposed pulp grafting technique in regenerating the bony defect within a short timeframe, as observed by the radiographic bone density measurements closely resembling those of the original bone after 4 months of follow-up. Consequently, the pulp graft demonstrates promising potential for achieving satisfactory alveolar bone repair within a relatively short period.

Conclusions

We conclude within the limits of this study

The effectiveness of dental pulp tissue in repairing alveolar bone defects in the maxilla was demonstrated. The effectiveness of the method which used in preparing the pulp graft and applying it in the bony defect. The described technique is simple, minimally invasive, and readily applicable in clinical practice without requiring complex in vitro procedures. The encouraging results obtained in this study warrant further investigations into the potential of this technique for repairing various types of bony defects in the facial and jaw regions.

Ethical approval

The approval of the ethics committee for research was obtained. The protocol of the study had been approved by the ethics committee of the college of Dentistry Research Centre at the university under approval (1362) during session (6).

Consent

Written informed consent was obtained from the patient for publication and any accompanying images. A copy of the written consent is available for review by the Editor-in-Chief of this journal on request.

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Not applicable.

Author contribution

All authors have approved the final draft of the manuscript.

Conflicts of interest disclosure

The authors declare no conflict of interest, financial or otherwise.

Research registration unique identifying number (UIN)

<https://www.researchregistry.com/browse-the-registry#userresearchregistry/registerresearchdetails/651340388f644a002889c5e4/researchregistry9558>.

Guarantor

Omar Aljarmakani.

Data availability statement

All data and material collected during this study are available from the corresponding author upon reasonable request.

Provenance and peer review

Not commissioned, externally peer-reviewed.

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