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Mandibular Bone Regeneration Following Maxillofacial Surgical Defects

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Abstract

This study aimed to quantitatively evaluate mandibular bone regeneration in veterinary rabbit models following experimentally induced maxillofacial defects using clinical and numerical healing parameters. An experimental in vivo study was conducted using 24 healthy adult rabbits randomly divided into three groups: control group, bone graft group, and platelet-rich fibrin (PRF) group. Standardized mandibular bone defects measuring 5 mm in diameter were surgically created under general anesthesia. Clinical healing evaluation, inflammatory scoring, and quantitative bone regeneration measurements were recorded at 2, 4, and 8 weeks postoperatively. Statistical analysis was performed using repeated-measures ANOVA and Tukey post hoc tests with significance set at $p < 0.05$. PRF group had significantly higher bone healing scores and better quantitative bone regeneration values than control group ($p < 0.05$). The healing results were also better in the bone graft group, but not as significant as in the PRF group. All groups experienced a gradual decrease in inflammatory scores during the experiment. Experimental mandibular defects created in rabbits showed successful regeneration of the bone. PRF demonstrated better regenerative effect and healing results than the traditional healing process. The quantitative clinical assessment methods were able to evaluate the progression of bone healing without a histological evaluation.

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Introduction

Regeneration of bone in the craniofacial region continues to be a formidable issue in oral and maxillofacial surgery. Mandibular defects can be caused by trauma, infection, tumor surgery, congenital abnormalities, or surgery, and often cause functional and esthetic complications. This is why it is crucial for the reconstruction of mandibular defects to restore the function of mastication, speech, facial symmetry and general oral function.

A key aspect of the development and assessment of regenerative surgical techniques is the use of experimental animal models prior to their clinical use in humans. Rabbits, as one of the laboratory animals, have been selected as highly suitable animals for maxillofacial surgical research for several reasons, such as rapid bone turnover, good healing properties, manageable size and relatively low maintenance [1]. Mandibular bone in rabbits has also been shown to have structural and biological similarity, which enable the processes of bone regeneration to be meaningfully evaluated.

Healing of mandibular defects is a complex process consisting of inflammatory, proliferative, and remodeling stages with several cellular and biochemical pathways involved. Inadequate vascularization, osteogenic cell activity, and scaffold stability are necessary for successful bone formation, which might lead to the formation of fibrous tissues, decreased mechanical stability, and poor functional recovery in case of delayed or inadequate bone healing.

Different methods using regenerative materials have been studied to improve the healing of the mandible such as autogenous bone grafts, synthetic materials, growth factors, stem cells and platelet concentrates. Platelet

rich fibrin (PRF) has recently garnered considerable interest due to its ease of preparation, autologous nature and capacity for growth factor release and stimulation of angiogenesis and osteogenesis [2]. PRF consists of platelets, leukocytes, cytokines and components of the fibrin matrix, which can promote tissue healing and enhance the regeneration outcome.

Bone grafting procedures continue to be one of the most frequently utilized methods to reconstruct maxillofacial defects. Bone graft materials are used to provide structure and serve as scaffolds for new bone growth. Some clinical issues such as graft resorption, donor site morbidity, and differences in healing remain to be addressed, however, which is why finding regenerative techniques that enhance healing remains an important goal in oral and maxillofacial surgery [3].

Histological and radiographic methods have been used in several studies to determine the results of bone regeneration. Histological analysis gives the detailed information at microscopic level but requires sacrifice of animal and the processing of the tissues for long. Quantitative clinical evaluation methods can provide a simpler, more reproducible assessment of the progression of bone healing, and decrease the complexity of the procedure [4].

Clinical parameters like healing scores, inflammatory response, defect closure measurements and bone density values can help to gain information on the regenerative performance. It can also be used to quantify the data, enabling easy comparison between treatment groups and a more objective assessment of healing as time passes.

Although there have been several advances in regenerative surgery, little research has been done in a veterinary rabbit model, where only quantitative and clinical analysis methods were used without histological evaluation. The purpose of the present study, therefore, was to quantify the regeneration of the mandible after experimental maxillofacial defects in rabbit models using clinical healing parameters and numerical measurements of the regenerate.

Materials and Methods

Study Design

In the present experimental in vivo study, the authors examined the rabbit model for maxillofacial bone defects in the mandible that were generated surgically to assess the bone regeneration potential.

Animal Selection

Twenty-four healthy male adult rabbits weighing 2.0 to 2.5 kg were used in the study. The animals were kept under standard laboratory conditions with controlled temperature, humidity and light cycles. Both rabbits in the experimental groups were fed the standard laboratory diet and supplied water ad libitum, throughout the experimental period.

Experimental Groups

The animals were randomly split into three groups of 8, with the same number of animals in each group.

- Group I: Control group
- Group II: Bone graft group
- Group III: Platelet-rich fibrin (PRF) group

Surgical Procedure

The surgical methods were all done under general anesthesia with the use of ketamine hydrochloride and xylazine. After the preparation and disinfection of the surgical site, the standardized circular defects with 5 mm diameters were made in the body of the mandibles with surgical round burs under continuous saline irrigation.

The defects in the control group were not treated and left to heal naturally. Bone grafts were performed in the bone graft group, using a synthetic bone graft material. In the PRF group, the PRF membranes were prepared and applied into the defects immediately following surgery.

Wound was closed with resorbable sutures and patients were treated with antibiotics and analgesics for five days.

In preparation of A-PRF, venous blood samples were taken from the patients in a sterile tube free of anti-coagulants and were centrifuged at 3000 rpm for 10 minutes. The resulting clot of fibrin was isolated and immediately inserted into the defects in the mandibles.

Clinical Evaluation

Assessment of clinical healing was made at 2, 4, and 8 weeks after surgery. All the experimental groups were evaluated carefully for postoperative swelling, inflammatory response, wound healing score, defect closure percentage and signs of infection for every evaluation period.

Quantitative Bone Regeneration Assessment

Bone regeneration was quantitatively assessed by standardized, numerical measurements derived from clinical defect assessment. Healing percentage and scores were determined at each evaluation time for each group, and the results were termed as the defect healing percentage and regenerative scores.

Statistical Analysis

The data were analyzed with the SPSS software 27. All quantitative variables were summarized as mean and standard deviation. Groups were compared and observation periods were compared using repeated-measures ANOVA with Tukey post hoc analysis. A p-value of < 0.05 was used for a statistical significance level.

Ethical Approval

All experimental procedures were done in accordance with the institutional guidelines for animal care with the approval of the research ethics committee of the Gilgamesh University, College of Dentistry (2025/17).

Results

Clinical Healing Evaluation

The surgical procedures were performed without significant complications in all animals in the postoperative period. Mild postoperative swelling was observed during the first week in all experimental groups, with gradual reduction over time. The PRF group demonstrated faster clinical healing and lower inflammatory scores compared with the control group.

The PRF group demonstrated significantly higher healing scores at all observation periods compared with the control group ($p < 0.05$).

Defect Closure Percentage

Quantitative assessment of mandibular defect closure demonstrated progressive healing in all groups throughout the experimental period. The PRF group showed the greatest percentage of defect closure, followed by the bone graft group.

Statistically significant differences were observed among the groups at all time intervals ($p < 0.05$).

Inflammatory Response Assessment

Postoperative inflammatory response gradually decreased throughout the healing period in all experimental groups. The control group demonstrated higher inflammatory scores compared with the treated groups.

The PRF group demonstrated significantly lower inflammatory scores compared with the control group during all healing intervals.

Quantitative Bone Regeneration Scores

Quantitative regenerative analysis demonstrated superior bone healing outcomes in the PRF-treated group. Progressive improvement in regenerative scores was observed over time in all groups.

Repeated-measures ANOVA revealed statistically significant improvements over time in all groups ($p < 0.05$).

Progressive improvement in quantitative bone regeneration scores was observed in all experimental groups throughout the healing period. The PRF group demonstrated superior regenerative performance compared with the bone graft and control groups at all observation intervals.

Discussion

Mandibular bone regeneration is an important challenge in oral and maxillofacial surgery especially in traumatic defects, cystic lesions, tumor resection or congenital

defects. Experimental animal studies are contributing to the understanding of the mechanisms and strategies for bone healing before these can be applied to humans. The model developed in this study was used in a rabbit for quantitative assessment of the regeneration of the mandibular bone in surgical defect of the maxillofacial region.

The rapid bone metabolism rate, excellent healing ability and manageable anatomical features make rabbits an ideal experimental animal for maxillofacial research. Rabbits have been shown in other studies to have reproducible and reliable mandibular defects for the testing of bone regenerative materials and healing response [5].

The results of the present study revealed progressive improvement in healing in all the experimental groups with the passage of time. Yet, the bone graft and PRF groups showed much more favorable regenerate than the untreated control group. These results indicate that regenerative biomaterials can positively affect the healing of mandibular defects and enhance the regeneration of bone.

The PRF group showed the best healing ability and the highest percentages of defect closure when compared to all groups tested. PRF has been in the spotlight in regenerative surgery due to the release of growth factors such as platelet-derived growth factor, transforming growth factor-beta, and vascular endothelial growth factor. These biological mediators are involved in angiogenesis, proliferation of osteoblasts, collagen synthesis and tissue remodeling [6].

Other possible reasons for the improvements in healing seen in the PRF group may be due to the fibrin matrix, which creates a biological scaffolding for cellular migration and tissue regeneration. Additionally, leukocytes found in PRF can play a role in antimicrobial and inflammatory response modulation in healing.

The same was observed in the bone graft group than the control group. The role of bone graft products is to support bone and act as an osteoconductive scaffold to promote new bone formation and stabilization of the defect. Regenerative performance was slightly reduced, however in comparison to the PRF group, this could be attributed to reduced biological integration and remodeling. In all groups, the inflammatory response was gradually reduced while healing. The inflammatory scores were low in the PRF group, representing the best biological compatibility and tissue healing. Minimizing post-operative inflammation can speed soft tissue healing and improve the overall regeneration.

One of the big benefits of this study was the use of quantitative clinical assessment methods. Quantitative evaluation permits multiple observations of healing without extensive tissue processing or sacrifices of the animal, as with histological evaluation. Regenerative data from clinical healing scores, inflammation, and defect closure measurements were reproducible and comparable.

Measuring bone regeneration in an experimental model has been emphasized by several previous investigations to be a combination of clinical and quantitative measurements. The quantitative analysis provides further objectivity and allows for comparison of various regenerative materials and surgical procedures [7].

While the results of this investigation were favorable, there are some limitations to be recognized. The study was conducted under the conditions of controlled experimental conditions which do not necessarily reflect the complexity of the defects in a maxillofacial area in real clinical practice. Further, the sample size was relatively small, and remodeling that lasted longer than eight weeks was not assessed.

Larger sample sizes, longer observation periods and more sophisticated regenerative biomaterials and biomimicking scaffolds could shed further light on mandibular bone regeneration in veterinary experimental models in the future.

Conclusion

The limitation of this experimental study, the results revealed that the surgically induced maxillofacial defects in the rabbit were successfully regenerated in the mandible. Bone graft treatment and platelet-rich fibrin (PRF) treatment both had significant improvement in healing compared with natural healing conditions. The PRF group had the best regenerative properties with higher QHS and a decreased inflammatory response and improved defect closure.

Based on the results of the present study, the use of platelet-rich fibrin as a regenerative method in enhancing bone healing in the mandible during maxillofacial surgery may be considered. Besides, quantitative clinical evaluation methods were reliable and reproducible for assessing bone regeneration without the need for histological analysis or radiographs.

Study Limitations

Although the experimental design and surgery in the present study were standardized, some limitations must be highlighted. This study was limited in scope of sample size and observation periods. Moreover, only quantitative clinical parameters were analyzed to keep the procedure as simple and reproducible as possible. Further research with longer follow-up periods and other forms of regenerative assessments could yield more information on the healing of mandibular bone in veterinary experimental models.

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Table 1. Clinical healing scores at different observation periods.

Group	2 Weeks Mean ± SD	4 Weeks Mean ± SD	8 Weeks Mean ± SD	p-value
Control Group	2.8 ± 0.5	4.6 ± 0.6	6.2 ± 0.7	<0.001
Bone Graft Group	4.1 ± 0.4	6.5 ± 0.5	8.3 ± 0.4	<0.001
PRF Group	4.8 ± 0.3	7.4 ± 0.4	9.1 ± 0.3	<0.001

Table 2. Percentage of mandibular defect closure.

Group	2 Weeks (%)	4 Weeks (%)	8 Weeks (%)	Percentage Improvement
Control Group	24.5 ± 3.2	48.6 ± 4.1	67.4 ± 5.0	175%
Bone Graft Group	38.7 ± 3.8	65.2 ± 4.4	86.9 ± 3.9	124%
PRF Group	45.8 ± 4.0	74.6 ± 3.7	93.5 ± 2.8	104%

Table 3. Postoperative inflammatory scores.

Group	2 Weeks Mean ± SD	4 Weeks Mean ± SD	8 Weeks Mean ± SD	Inflammatory Grade
Control Group	3.9 ± 0.5	2.8 ± 0.4	1.9 ± 0.3	Moderate
Bone Graft Group	3.1 ± 0.4	2.0 ± 0.3	1.2 ± 0.2	Mild
PRF Group	2.4 ± 0.3	1.5 ± 0.2	0.8 ± 0.1	Minimal

Table 4. Quantitative bone regeneration scores.

Group	2 Weeks Mean ± SD	4 Weeks Mean ± SD	8 Weeks Mean ± SD	p-value
Control Group	3.5 ± 0.6	5.2 ± 0.5	6.8 ± 0.7	<0.001
Bone Graft Group	5.1 ± 0.5	7.0 ± 0.6	8.5 ± 0.5	<0.001
PRF Group	5.8 ± 0.4	8.1 ± 0.5	9.4 ± 0.3	<0.001

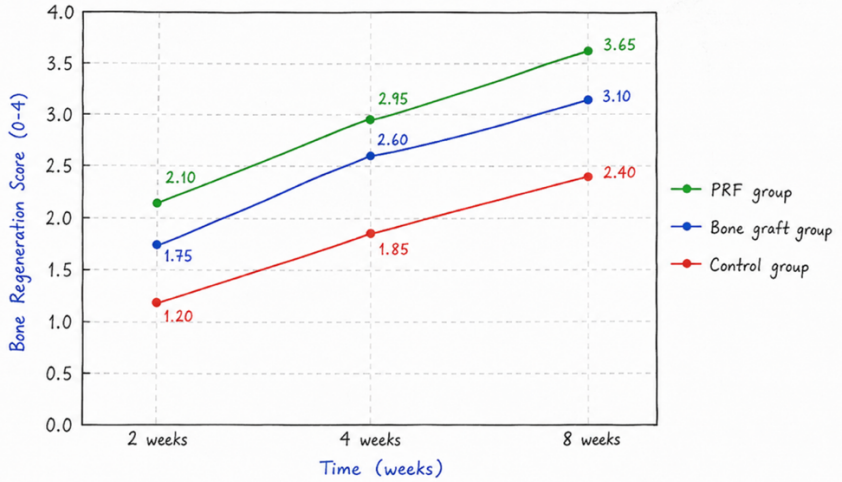

Figure 1. Comparative quantitative bone regeneration scores among experimental groups during the healing period.