Journal of International Oral Health 2016; 8(7):801-807

Received: 17th February 2016 Accepted: 10th May 2016 Conflict of Interest: None

Source of Support: The Vice Chancellor for Research of Tabriz University of Medical Sciences

Original Research

Doi: 10.2047/jioh-08-07-11

Evaluation of the Effect of Low-level Laser on Biomaterials Used in Maxillary Sinus Grafts Using Histological and Radiologic Examinations: A Randomized Controlled Clinical Trial

Mohammad-Taghi Chitsazi[1], Leila Roshangar[2], Adileh Shirmohammadi[1], Farzad Esmaieli[3], Ali-Hossein Dehghani[4], Mahdi Rahbar[5], Masoumeh Faramarzi[6]

Contributors:
[1]Professor, Department of Periodontics, Tabriz Dental Faculty, Tabriz University of Medical Sciences, Tabriz, Iran; [2]Professor, Department of Histology, Medical School, Tabriz University of Medical Sciences, Tabriz, Iran; [3]Associate Professor, Department of Dental and Maxillofacial Radiology, Tabriz Dental Faculty, Tabriz University of Medical Sciences, Tabriz, Iran; [4]Post-graduate Student, Department of Periodontics, Tabriz Dental Faculty, Tabriz University of Medical Sciences, Tabriz, Iran; [5]Postgraduate Student, Department of Operative and Esthetic Dentistry, Faculty of Dentistry, Tabriz University of Medical Sciences, Tabriz, Iran; [6]Associate Professor, Department of Periodontics, Dental and Periodontal Research Center, Tabriz University of Medical Sciences, Tabriz, Iran.

Correspondence:
Dehghani A. Department of Periodontics, Tabriz Dental Faculty, Golgasht St., Tabriz, Iran. Tel.: +984113355965 (Extension: 232), Phone: +989143110538, Fax: +984113346977. Email: a_h_dehgani@yahoo.com

How to cite the article:
Chitsazi M, Roshangar L, Shirmohammadi A, Esmaieli F, Dehghani A, Rahbar M, Faramarzi M. Evaluation of the effect of low-level laser on biomaterials used in maxillary sinus grafts using histological and radiologic examinations: A randomized controlled clinical trial. J Int Oral Health 2016;8(7):801-807.

Abstract:

Background: One of the techniques suggested for the provision of adequate bone in the maxillary sinus area for placement of endosteal implants with a standard length is to use the sinus lift surgical technique along with autogenous bone graft or bone substitute materials. The aim of this study was an evaluation of the effect of low-level laser on graft materials used for augmentation of the maxillary sinus.

Materials and Methods: In the present randomized clinical trial with a split-mouth design, 19 patients aged 30-80 years were evaluated. All the subjects underwent a bimaxillary sinus lift surgical procedure. In the control group, freeze-dried bone allograft (FDBA) and platelet-rich fibrin (PRF) allograft materials were used; in the case group, in addition to the materials mentioned above, low-level 980-nm diode laser beams were applied. Six months after surgery, the density of bone in the augmented area was determined with the cone-beam computed tomography technique.

Results: The results showed that the percentages of bone formed in the control and case groups were 20.10 ± 5.67% and 36.26 ± 11.26%, respectively, with a significant difference between the two groups ($P < 0.05$). There was no significant difference in the means of connective tissue between the two groups ($P = 0.612$). The Hounsfield unit exhibited a significant difference in relation to the mean bone density within the graft material between the two groups ($P = 0.000$).

Conclusion: Low-level 980-nm diode laser beams can improve the results of treatment rendered with the use of FDBA and PRF.

Key Words: Bone graft materials, low-level laser, maxillary sinus augmentation, platelet-rich fibrin

Introduction

Use of endosteal dental implants has some limitations in many cases due to some anatomical problems or the atrophy of the alveolar crest. One of these problems is the maxillary sinus in the posterior maxilla. In this area, due to the extraction of teeth, transverse and vertically resorption of the alveolar bone, and pneumatization of the sinus, in many cases, sufficient amount of bone does not exist for the placement of endosteal implants.[1,2] In most clinical situations, the bone in this area has low quality and based on studies by Misch use of short implants (<10 mm in length) in the posterior maxilla has the highest failure rate.[2,3]

One of the techniques suggested for the provision of adequate bone in that area for the placement endosteal implants with a standard length is sinus lift surgical procedure in association with autogenous bone graft or bone substitute materials. Recent advances in surgical techniques, implants, and grafts materials have improved the prognosis of implant treatments in the posterior maxilla. Autogenous bone grafts have been considered the gold standard for a long time due to their osteogenic potential; however, these grafts have some disadvantages.[4,5]

Despite the fact that autogenous bone has osteoconductive, osteoinductive, and osteogenic properties, it is a suboptimal choice due to its high resorption rate, limited intra- and extra-oral sources for harvesting bone, longer and more numerous surgical steps, and complications associated with the donor site.[5] On the other hand, use of bone substitute materials (allografts, xenografts, and alloplasts) and various clinical reports on their clinical results seems to be logical.[5-7]

Currently, allograft materials that predominantly exhibit osteoconductive properties and provide the required scaffold for the proliferation of and osteogenesis by osteoblasts have attracted attention for sinus lift procedures in association with platelet-rich fibrin (PRF), which is a rich source for the different growth factors. PRF as the second generation of platelet

concentrate results in the release of platelet-derived growth factor (PDGF) and transferring growth factor (TGF). PDGF is a glycoprotein with mitogenic and angiogenic properties and can stimulate macrophages. TGF has chemotactic properties and can stimulate osteoblast progenitors to differentiate into mature osteoblasts. Furthermore, it can inhibit differentiation of osteoclasts and resorption of bone.[8-10]

One of the problems in allografts is the slow integration of the graft material with the host site (graft-host interface). To solve these problems, growth factors and low-level lasers (LLLs) are used.[8-11] LLL accelerate bone generation and wound healing by exerting their effect on mitochondria, production of large amounts of ATP, angiogenesis and by increasing proliferation of osteoblasts, fibroblasts, macrophages, etc.[12-14] Various *in vivo* and *in vitro* studies have evaluated the photobiostimulatory effects of laser and have reported various results in terms of the laser type, the effects studied, and the tissue in question.[3,15] Considering the safety of LLLs and their ability to accelerate cellular activity and considering the fact that PRF is an autogenous agent rich in different growth factors and is an allograft with good osteoconductive properties, the present study was designed to answer the question whether LLL can improve the quality and quantity of the bone formed.

Materials and Methods

Patient selection and study design

The present study had a split-mouth design, in which 19 patients 40-80 years of age with Kennedy Cl I partial edentulism or complete edentulism in the maxilla were evaluated. To determine the sample size, first five patients underwent treatment in a pilot study. In one group, the mean percentage of bone formed was 31.90 ± 10.64%, and in the other group, it was 26.80 ± 5.89%. The sample size was estimated at 19 patients at $\alpha = 0.05$ and a study power of 80% with a difference of 8 units.[1]

The inclusion criteria consisted of Kennedy Cl I partial edentulism (bilateral edentulism) or complete edentulism in the upper jaw, a residual bone height <4 mm between the alveolar crest and the maxillary sinus floor, absence of systemic diseases, no smoking habits, no pregnancy at the time of the study, absence of any pathologic conditions in the maxillary sinus, absence of any untreated periodontal diseases and periapical pathoses, and a normal platelet count.

Preparation of PRF

PRF was prepared based on a protocol used by Dohan *et al.*[16] A total of 10 mL of blood was placed in sterilized tubes without any anticoagulant. The tubes were centrifuged at 2700 rpm in a single stage for 10 min. After centrifugation, three layers were distinguishable within the tubes: The lowermost layer was the RBC layer; the middle layer was the fibrin lot layer, and the uppermost layer was acellular plasma layer. The uppermost layer was discarded, and the middle and lowermost layers were separated (Figure 1a).

The surgical technique

About 1 h before the surgical procedure, the patients' medical and dental histories were reviewed, and a CBC test was ordered. Then, 400 g of ibuprofen and 625 mg of coamoxiclav were given to each patient. The patients rinsed their oral cavity with 0.2% chlorhexidine gluconate solution for 1 min before surgery. Then, the surgical site was anesthetized with 2% lidocaine (3.6 mL). To gain access to the maxillary sinus, first, a transverse incision was made on the alveolar ridge crest and two vertical incisions were made anterior and posterior to the transverse incision, and a full-thickness flap was elevated.[17] Then, a piezosurgery round diamond bur (NSK, Model NE214) was used to remove a bony window, measuring approximately 15-20 mm^2, under saline solution irrigation on the lateral wall of the sinus. Then, the sinus membrane was carefully elevated so as not to perforate it (Figure 1b). Then, freeze-dried bone allograft (FDBA), with a granule size of 750-110 nm, was mixed with fibrin clot and placed on the sinus floor (Figure 1c).

Laser irradiation

After the completion of surgical and augmentation procedures, LLL beams (diode laser with a power of 0.5 W, Biolase, Germany) at a wavelength of 98-nm and beam energy of 10 J/cm were applied on one side randomly based on the study protocol for 20-30 s 3 times each week every other day. The laser-irradiated group was assigned to the case (intervention)

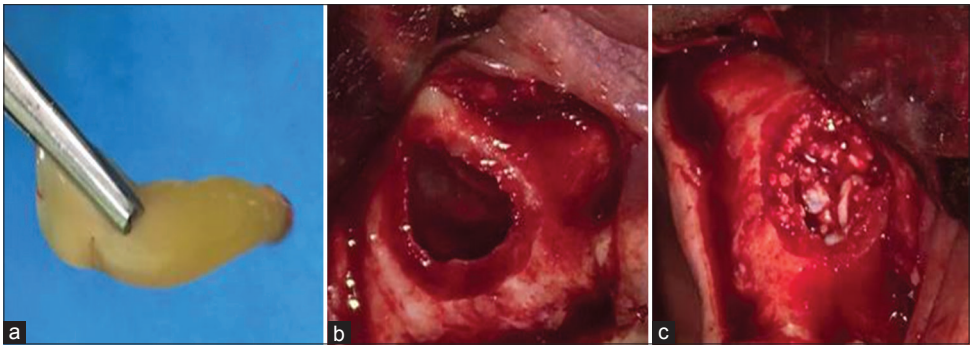

Figure 1: (a) The intermediate layer (the fibrin clot) which is ready for being mixed with freeze-dried bone allograft, (b) preparation of an osseous window in the lateral wall, and (c) placement of the mixture of platelet-rich fibrin and allograft within the sinus.

group, and the contralateral side was assigned to the control group. Since no specific protocol is available in this respect, in the majority of studies, the number of sessions and the duration of irradiation have been selected based on the researchers' opinions.[18-20]

Histological examination

To place the implants after 6 months in both groups, first, the osteotomy area was determined and then a thin trephine bur, measuring 2.7 mm in internal diameter, was used to collect histological samples from the osteotomy area. The biopsy samples were placed in coded bottles containing 10% formation at pH = 7 and sent to the laboratory for histological evaluation. Then, the samples were fixed in 10% formalin for 10 days and then decalcified in 65% nitric acid for 72 h. Subsequently, the samples were once again placed in buffered formalin for 1 week and then rinsed under running water.

The samples were dehydrated in increasing concentrations of ethanol for one hour and then washed twice with xylene. Then, they were embedded in paraffin and cut to a thickness of 5-6 μm by a microtome (Figure 2). The specimens were stained with hematoxylin and eosin using a standard technique and prepared for histological analysis.

The parameters were evaluated in terms of the percentage of newly formed bone, the residual graft materials, and connective tissue under a light microscope (BX40, Olympus, Germany) (Figure 2). In addition, the images were analyzed using Notice Software Image computer program.[18]

Radiographic evaluation

After the surgery, to evaluate the area for implant placement, that area underwent a cone-beam computed tomography (CBCT) examination. The CBCT mages were used to compare the distance between the alveolar crest and the sinus floor before surgery and 6 months after surgery. In addition, the Hounsfield unit (HU) was used to evaluate the density of the graft materials from the sinus floor up to the ridge crest at baseline and 6 months after sinus lift surgery with NNT Viewer 2 software program.

Statistical analysis

Kolmogorov–Smirnov test was used to evaluate the normal distribution of the variables under study. Due to the normal distribution of all the data, descriptive statistics (means, standard deviations, frequencies, and parcentages), mean comparison test and *t*-test were used for statistical analysis of data. Statistical significance was set at $P < 0.05$.

Results

Kolmogorov–Smirnov test showed normal distribution of data of the variables. All the variables were over 0.05 (Table 1). Paired *t*-test showed that the mean percentages of bone formed in the control (allograft + PRF) and case (allograft + PRF + LLL) groups were 20.10 ± 5.67% and 36.26 ± 11.62%, respectively, with significantly higher percentage of bone formed in the case group ($P < 0.05$). In addition, paired *t*-test showed that the mean percentages of residual graft materials in the control and case groups were 29.11 ± 11.79% and 24.82 ± 10.09%, respectively, with no statistically significant differences. In other words, there was a similar amount of residual graft materials in the control and case groups ($P = 0.330$). On the other hand, the percentages of connective tissue in the control and case groups were 44.640 ± 16.46% and 41.40 ± 21.06%, respectively, with no significant differences between the two groups ($P = 0.612$).

In addition to histological evaluations, the results of CBCT examinations showed that the mean bone densities within the graft materials in the control and cases group in terms of the HU were 296.79 ± 37.55 and 339.16 ± 46.43, respectively,

Table 1: Distribution of data of the variables evaluated.

Variables	*n*	Control		Case	
		Kolmogorov–Smirnov Z	*P*	Kolmogorov–Smirnov Z	*P*
Newly formed bone	19	1.04	0.223	0.770	0.593
Residual connective tissue	19	0.985	0.287	0.854	0.459
Connective tissue	19	0.619	0.839	0.720	0.678
Bone density	19	0.565	0.907	1.271	0.079
The height of the maxillary sinus floor	19	0.998	0.273	0.817	0.522

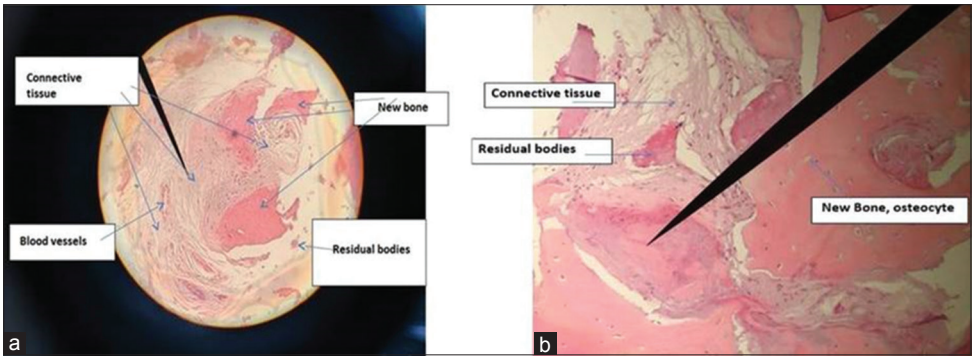

Figure 2: (a and b) Histologic analysis: Newly formed bone, the residual graft materials, and connective tissue under a light microscope (the specimens were stained with hematoxylin and eosin using a standard technique and magnification was ×400).

with significant differences between the two groups ($P = 0.000$) (Table 2).

Comparison of the distance between the sinus floor and the crest of the alveolar ridge before and after therapeutic intervention showed that before intervention, the distances were 3.678 ± 7.02 and 3.584 ± 0.673 mm in the control and case groups, respectively, with no significant differences ($P = 0.402$). Six months after intervention, these values increased to 11.252 ± 0.589 and 12.094 ± 0.712 mm in the control and case groups, respectively, with a significant difference between the two groups ($P = 0.002$) (Table 3 and Figure 3).

Table 2: The differences in mean percentages of newly formed bone, residual graft materials, the amount of connective tissue and bone density between the case and control groups.

Variables	*n*	Mean	SD	Paired-sample *t*-test		
				t	df	*P*
Amount of bone						
Control	19	20.105	5.675	−5.98	18	0.000
Case	19	36.263	11.627			
Percentage of residual graft material						
Control	19	29.11	11.798	1.57	18	0.330
Case	19	24.82	10.092			
Amount of connective tissue						
Control	19	44.646	16.46	0.516	18	0.612
Case	19	41.408	21.064			
Bone						
Control	19	296.79	37.55	−5.200	18	0.000
Case	19	339.16	46.43			

df: Degree of freedom, SD: Standard deviation

Table 3: Comparison of the distance between the sinus floor and the alveolar ridge crest in the case and control groups before intervention and 6 months after intervention.

Time	*n*	Mean	SD	Paired-sample *t*-test		
				t	df	*P*
Baseline						
Control	19	3.6789	0.70204	0.858	18	0.402
Case	19	3.5842	0.67352			
After 6 months						
Control	19	11.2526	0.85918	−3.642	18	0.002
Case	19	12.0947	0.71217			

df: Degree of freedom, SD: Standard deviation

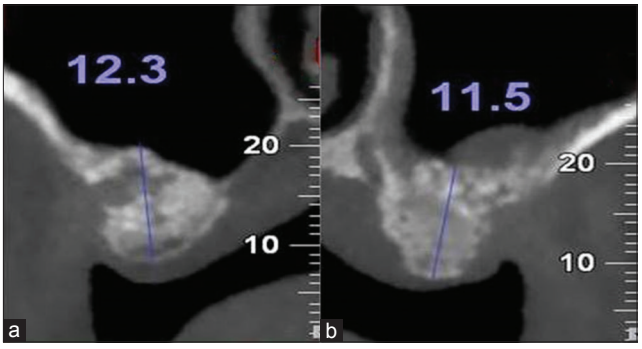

Figure 3: Cone-beam computed tomography image after graft procedure in the case group (a) and the control group (b).

Discussion

Augmentation procedures are required in the posterior maxilla for placement of implants in many cases. The most commonly used treatment modality in this area consists of elevating the sinus membrane and placement of a graft material beneath this membrane. Various studies have been carried out to identify an effective material which is inexpensive at the same time, and different results have been achieved.[12,18-22] Although surgical techniques, the histological and clinical results, use of different materials, timing of implant placement, and the stability of implants have been evaluated,[17,18,22] the present study evaluated the effect of LLL beams on the histological and radiologic views after placement of a combination of FDBA and PRF on the sinus floor during sinus floor augmentation procedures.

In the present study, the mean percentage of bone formed in the central group (PRF/FDBA) was 20.10 ± 5.67%. Consistent with the results of the present study, Dohan *et al.* reported that the percentage of newly formed bone in the PRF/FDBA group was 20.95%. Zhang *et al.* reported a mean percentage of 18.35 ± 6.62% for newly formed bone in the deproteinized bovine bone mineral/PRP group.[16,23-26] In contrast, Kolerman *et al.* reported a percentage of 1.29% for newly formed bone. Mazor *et al.* carried out a case series study, in which they used PRF alone and placed implants simultaneously, reporting a percentage of 93 ± 5% for bone formation around implants. Xuan *et al.* carried out an animal study and reported 41.8 ± 5.9% of bone formation in the PRF/Bio-Oss group.[23-25] Although there are differences between the two latter studies and the present study in relation to the percentage of newly formed bone, there is indication that PRF alone or in combination with graft materials and bone substitutes is effective information of new bone and exhibits some degrees of success, which might be attributed to the presence of different growth factors in the complex fibrin plexuses in PRF; these growth factors include PDGF, TGF-β1, β2, vascular endothelial growth factors, platelet-activating factor 4, (platelet-derived endothelial growth factors), interleukin 1 and 2, and basic fibroblast growth factor. Considering the advantages and capability of PRF when it is mixed with other bone substitutes in achieving the relative aims of the clinician in treatment results and infliction of no extra costs on the patient and since the selected bone samples were <4 mm between the crest of the alveolar ridge and the sinus floor; in the present study, we had no other choice but to mix PRF with other bone substitutes such as FDBA. In addition, in this context, we considered the fact that due to the initial instability of the implant it was not possible to carry out augmentation and place the implants at the same time; furthermore, RRF has a weak scaffold and is resorbed gradually after surgery. Therefore, in the present study, the formation of bone cannot be attributed to PRF alone; in this context, the success of treatment is not the result of the use of FDBA alone. On the other hand, other studies have shown that when PRF is mixed with other bone substitutes, the success rate increases

and even when PRF has been used alone, it has exhibited a good success rate.[8,24]

In the present study, the amount of residual graft material in the control group was 29.11 ± 11.79%. Consistent with the results of the present study, Zhang *et al*. reported that the amount of residual graft material in the deproteinized bovine mineral/PRP group was 19.16 ± 6.89%.[26] In contrast, Dohan *et al*. reported that the amount of residual graft material in the PRF/allograft group was 9.41%.[16] Several studies have evaluated the amount of residual graft material in the augmented area, and different results have been achieved.[16,23] Although the principal aim of this study was not to evaluate the amount of residual graft material in the augmented area, it appears the resorbability, particle size, and the duration of persistence in the graft area affects the amount of residual graft material; in this context, it should be pointed out that the percentage of the residual graft material affects the osseointegration of the implant.

In the present study, the area under study was evaluated radiographically, in addition to histological evaluations. Six months after surgery, the area underwent CBCT examinations, which showed new bone formation in the augmented area, with a mean bone density of 296.79 ± 37.55 in terms of HU. In addition, the distance between the crest of the alveolar ridge and sinus floor in the control group was 3.678 ± 7.02 mm before surgery, which increased to 11.252 ± 0.859 mm 6 months after surgery. Tajima *et al.* reported a mean bone density of 323±156.2 HU for the newly formed bone. In this context, the mean bone height of 11.8 ± 1.67 mm might be evaluated relative to bone density in terms of HU.[26] HU is directly related to the tissue attenuation coefficient. This parameter is a relative scale defined for different types of bone, including cortical bone with a very high density (>600 HU), cortical bone along with cancellous bone with moderate density (400-600 HU), and cortical bone along with cancellous bone with low density (<200 HU).[26]

In the present study, the density of the newly formed bone in the graft area was comparable to that in the posterior maxilla. Therefore, it might be claimed that sinus augmentation with PRF/allograft combination is reliable for implant placement. In addition, the bone height (11.252 ± 0.859 mm) achieved by this graft was at an acceptable level, although the achieved height probably depends on the amount of elevation carried out for sinus membrane and the volume of the graft material used.

The principal aim of the present study was to evaluate the effect of LLL on the area grafted with PRF/allograft. LLL therapy is applied in dentistry for wound healing. On the other hand, the majority of studies on the effects of LLL have been carried out on the healing of skin wounds.[27] These studies have shown that LLL has some effects on the wound including an increase in cellular metabolic processes, an increase in the regeneration potential of biologic tissues, an increase in neoangiogenesis and formation of regenerative tissues.[28] Different animal studies have shown that application of LLL, in association with PRP, results in the acceleration of wound healing and promotion of osteogenesis in fenestrations in the alveolar bone.[29,30] The mechanism of action of LLLs has not been completely elucidated. However, it appears its nature is photochemical and is related with an increase in cellular proliferations through photochemical changes in intercellular chromatic molecules in mitochondria. On the other hand, this mechanism is multifactorial and includes establishment of angiogenesis, collagen synthesis and maturation, and revival of mitochondria and osteogenic cells by LLLs, too.[27]

In the present study, histological evaluation in the case group (PRF/allograft/LLL) showed that the amount of newly formed bone was 36.26 ± 11.62%; however, the amount of newly formed bone in the control group was 20.10 ± 5.67%, with statistically significant differences between the two groups, despite the limited number of samples ($P < 0.05$). In addition, the amount of residual connective tissue in the case group was 41.4 ± 21.06%, which was less than that in the control group (44.640 ± 16.46%), but the difference was not significant. However, CBCT examinations showed that the mean bone density within the graft material in the control group was 296.79 ± 37.55 HU, which was less than that (339.16 ± 46.43 HU) in the case group, and such difference was statistically significant ($P = 0.000$). Since the amount of newly formed bone is an important parameter in the surgical augmentation of maxillary sinus, an increase in the amount of bone formed in the present study might be attributed to the presence of various growth factors in the complex fibrin plexuses in PRF and the accelerating effect of LLL on increasing metabolism, promotion of angiogenesis, and synthesis of collagen, which result in formation of more bone.

Differences in the parameters evaluated between the control and case groups possibly indicate the effect of laser on improving treatment results with the application of laser beams when PRF/allograft combination was used for sinus augmentation and might boost the hypothesis that LLL has synergistic therapeutic effects with PRF. However, since in the present study PRF was used in association with allograft, probably the effects of laser cannot be attributed to PRF alone and even if PRF had been used alone, given the effects of laser discussed above, it appears the study design, in which the effect of laser is shown on PRF would be rational. In addition, it seems it is rational to design an animal study, in which it would be possible to lift the sinus membrane with the use of no material to evaluate the effects of laser in that area; it might be possible in such a case to determine the positive therapeutic effects of PRF and laser.

Conclusion

Based on the results of statistical analyses, the amount of bone formed, the bone density, and the height of the sinus floor (the

crest of the residual ridge) were significantly greater in the case group compared to the control group after 6 months. However, the amounts of residual graft material and the connective tissue were similar in the case and control groups.

Acknowledgment
The authors wish to thanks The Vice Chancellor for Research of Tabriz Dental Faculty for their supports.

References

1. Barone A, Santini S, Sbordone L, Crespi R, Covani U. A clinical study of the outcomes and complications associated with maxillary sinus augmentation. Clin Oral Implants Res 2006;21:81-5.
2. Stavropoulos A, Karring T, Kostopoulos L. Fully vs. Partially rough implants in maxillary sinus floor augmentation: A randomized controlled clinical trial. Clin Oral Implants Res 2007;18(1):95-102.
3. Del Fabbro M, Testori T, Francetti L, Weinstein R. Systematic review of survival rates for implants placed in the grafted maxillary sinus. Int J Periodontics Restorative Dent 2004;24(6):565-77.
4. Fuentes R, Gonzalo O, Olate S, Rouco M, Borie E, Engelke W. Histological evaluation of bone regeneration means freeze dried bone allograft (FDBA) in post exodontia sockets. Int J Morphol 2011;29(1):286-92.
5. Gonshor A, McAllister BS, Wallace SS, Prasad H. Histologic and histomorphometric evaluation of an allograft stem cell-based matrix sinus augmentation procedure. Int J Oral Maxillofac Implants 2010;26(1):123-31.
6. Jensen OT, Shulman LB, Block MS, Iacono VJ. Report of the sinus consensus conference of 1996. Int J Oral Maxillofac Implants 1998;13 Suppl:11-45.
7. Mangano C, Bartolucci EG, Mazzocco C. A new porous hydroxyapatite for promotion of bone regeneration in maxillary sinus augmentation: Clinical and histologic study in humans. Int J Oral Maxillofac Implants 2003;18(1):23-30.
8. Butterfield KJ, Bennett J, Gronowicz G, Adams D. Effect of platelet-rich plasma with autogenous bone graft for maxillary sinus augmentation in a rabbit model. J Oral Maxillofac Surg 2005;63(3):370-6.
9. Choi BH, Im CJ, Huh JY, Suh JJ, Lee SH. Effect of platelet-rich plasma on bone regeneration in autogenous bone graft. Int J Oral Maxillofac Surg 2004;33(1):56-9.
10. Dugrillon A, Eichler H, Kern S, Klüter H. Autologous concentrated platelet-rich plasma (cPRP) for local application in bone regeneration. Int J Oral Maxillofac Surg 2002;31(6):615-9.
11. Pinheiro AL, Limeira Júnior Fde A, Gerbi ME, Ramalho LM, Marzola C, Ponzi EA, *et al.* Effect of 830-nm laser light on the repair of bone defects grafted with inorganic bovine bone and decalcified cortical osseus membrane. J Clin Laser Med Surg 2003;21(5):301-6.
12. Pjetursson BE, Tan WC, Zwahlen M, Lang NP. A systematic review of the success of sinus floor elevation and survival of implants inserted in combination with sinus floor elevation. J Clin Periodontol 2008;35 Suppl 8:216-40.
13. Torres CS, dos Santos JN, Monteiro JS, Amorim PG, Pinheiro AL. Does the use of laser photobiomodulation, bone morphogenetic proteins, and guided bone regeneration improve the outcome of autologous bone grafts? An *in vivo* study in a rodent model. Photomed Laser Surg 2008;26(4):371-7.
14. Yaakobi T, Maltz L, Oron U. Promotion of bone repair in the cortical bone of the tibia in rats by low energy laser (He-Ne) irradiation. Calcif Tissue Int 1996;59(4):297-300.
15. Geurs NC, Wang IC, Shulman LB, Jeffcoat MK. Retrospective radiographic analysis of sinus graft and implant placement procedures from the Academy of Osseointegration Consensus Conference on Sinus Grafts. Int J Periodontics Restorative Dent 2001;21(5):517-23.
16. Dohan DM, Choukroun J, Diss A, Dohan SL, Dohan AJ, Mouhyi J, *et al.* Platelet-rich fibrin (PRF): A second-generation platelet concentrate. Part I: Technological concepts and evolution. Oral Surg Oral Med Oral Pathol Oral Radiol Endod 2006;101(3): e37-44.
17. Boyne PJ, James RA. Grafting of the maxillary sinus floor with autogenous marrow and bone. J Oral Surg 1980;38(8):613-6.
18. Chen JW, Zhou YC. Effect of low level carbon dioxide laser radiation on biochemical metabolism of rabbit mandibular bone callus. Laser Ther 1989;2:83-7.
19. Karu TI. Mechanism of low-power laser light action on cellular level. In: Simunovic Z, (Editor). Laser in Medicine and Dentistry, Vita Graf Croatic, Croatia: Vitagraf; 2000.
20. Morimoto Y, Arai T, Kikuchi M, Nakajima S, Nakamura H. Effect of low-intensity argon laser irradiation on mitochondrial respiration. Lasers Surg Med 1994;15(2):191-9.
21. Dilsiz A, Canakci V, Aydin T. The combined use of Nd: YAG laser and enamel matrix proteins in the treatment of periodontal infrabony defects. J Periodontol 2010;81(10):1411-8.
22. Misch CE. Maxillary sinus augmentation for endosteal implants: Organized alternative treatment plans. Int J Oral Implantol 1987;4(2):49-58.
23. Kolerman R, Tal H, Moses O. Histomorphometric analysis of newly formed bone after maxillary sinus floor augmentation using ground cortical bone allograft and internal collagen membrane. J Periodontol 2008;79(11):2104-11.
24. Mazor Z, Horowitz RA, Del Corso M, Prasad HS, Rohrer MD, Dohan Ehrenfest DM. Sinus floor augmentation with simultaneous implant placement using Choukroun's platelet-rich fibrin as the sole grafting material: A radiologic and histologic study at 6 months. J Periodontol 2009;80(12):2056-64.
25. Xuan F, Lee CU, Son JS, Jeong SM, Choi BH. A comparative study of the regenerative effect of sinus bone grafting with platelet-rich fibrin-mixed Bio-Oss® and commercial fibrin-mixed Bio-Oss®: An experimental study. J Craniomaxillofac Surg 2014;42(4):e47-50.
26. Zhang Y, Tangl S, Huber CD, Lin Y, Qiu L, Rausch-Fan X. Effects of Choukroun's platelet-rich fibrin on bone

regeneration in combination with deproteinized bovine bone mineral in maxillary sinus augmentation: A histological and histomorphometric study. J Craniomaxillofac Surg 2012;40(4):321-8.
27. Tajima N, Ohba S, Sawase T, Asahina I. Evaluation of sinus floor augmentation with simultaneous implant placement using platelet-rich fibrin as sole grafting material. Int J Oral Maxillofac Implants 2013;28(1):77-83.
28. Almeida-Lopes L, Rigau J, Zângaro RA, Guidugli-Neto J, Jaeger MM. Comparison of the low level laser therapy effects on cultured human gingival fibroblasts proliferation using different irradiance and same fluence. Lasers Surg Med 2001;29(2):179-84.
29. El Khouly Castilla I. Maxillary Sinus Augmentation Using Growth Factors and Collagen Membranes [PhD Degree Thesis]: Universidad de Granada; 2014.
30. Seifi M, Atri F, Kaveh M, Yazdani M, Ebadifar A. Evaluation of low level laser effect, along with the application of DFDBA in rabbit bone repair. J Dent Fac Shahid Beheshti Univ Med Sci 2012;29(4):346-51.