



EFFICACY OF ANKAFERD BLOOD STOPPER® ON HEALING AFTER OSSEOUS GRAFTING IN THE TREATMENT OF PERIODONTAL INTRABONY DEFECTS

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ABSTRACT

Objective: Periodontal diseases are multifactorial infections that lead to destruction of the periodontal structures, including the tooth supporting tissues, periodontal ligament, and alveolar bone. The present study was designed to verify effect of Ankaferd Blood Stopper® (ABS) in conjunction with β tricalcium phosphate (β -TCP) on healing of periodontal intrabony defects. **Subjects and Methods:** A total of 20 intrabony periodontal defects were treated in ten patients with periodontal pocket depth (PPD) ≥ 6 mm and radiographic osseous defect depth ≥ 4 mm. The split mouth design was performed; In the 1st group (control group) ten defects were treated by β -TCP, while in the 2nd group (test group) ten defects were treated by mixing 0.5 ml of ABS with β -TCP. Phase I therapy was completed for all chosen patients to provide an oral environment more favorable for wound healing. Plaque index (PI), Gingival index (GI), Periodontal pocket depth (PPD) and clinical attachment level (CAL) were assessed for all patients. All indices were recorded at baseline, 6 and 12 months after surgery by the same operator. Radiographic measurements of bone density, M-D width of the defect and depth of the defect (DB) were also performed at the same time periods of recording the clinical parameters. **Results:** β -TCP/ABS group showed less mean plaque index, gingival index, clinical attachment loss, M-D width of the defect, depth of the defect and higher mean bone density than β -TCP group. **Conclusion:** Both treatment modalities demonstrated minimal inflammatory signs around teeth. Ankaferd Blood Stopper® combined with Beta tricalcium phosphate may enhance the healing and regenerative processes and lead to more bone fill, as well as density, compared to β tricalcium phosphate alone.

Keywords: Periodontal diseases, intrabony defects, bone grafting, Ankaferd Blood Stopper®.

INTRODUCTION

Periodontal diseases are chronic inflammatory disorders localized to the attachment structures of the teeth and considered to be the major cause of tooth loss in adults and the most prevalent form of bone pathology in humans⁽¹⁾. The term periodontal disease refers to both gingivitis and periodontitis.

Gingivitis is an inflammatory condition of the soft tissues surrounding the teeth. Periodontitis is an inflammatory disease resulting in inflammation of the supporting tissues, progressive attachment loss, and alveolar bone resorption⁽²⁾.

In conjunction with surgical access, bone or bone substitutes have been placed in the debrided

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periodontal defect with the aim of promoting periodontal regeneration. Several materials and techniques were used in the management of the intrabony defects, one of them is β tricalcium phosphate (β -TCP). Highly purified β -TCP has been shown to have osteoconductive activity and biodegradable nature in human bone⁽³⁾.

Ankaferd Blood Stopper® (ABS) has been recently introduced as a medicinal plant extract product and approved for the management of hemorrhage. The hemostatic effect of ABS is based on the formation of an encapsulated protein network that provides focal points for vital erythrocyte aggregation^(4,5). Ankaferd Blood Stopper® has been shown to affect endothelium, blood cells, angiogenesis, cellular proliferation, vascular dynamics and cell mediators as well as exerting antibacterial effects⁽⁶⁾. It enhances wound healing and accelerates early stage bone tissue formation. Moreover, it can decrease the occurrence of inflammation and necrosis, while increasing new bone formation during the early bone healing period⁽⁷⁾. Ankaferd Blood Stopper® accelerates bone healing and can be used alone or with collagenated heterologous bone graft in sinus floor augmentation procedures⁽⁸⁾.

Cone beam Computed tomography (CBCT) in dentistry has become popular and is commonly utilized for treatment planning of dental and periodontal problems. This imaging technique can allow the clinician to evaluate the condition of the patient's osseous structures in three dimensions without interferences of overlying structures. Three-dimensional imaging gives the clinician an ability to visualize and measure bone level without structures being superimposed on each other and has the potential to significantly enhance periodontal diagnosis compared to conventional radiographs⁽⁹⁾.

The present work was undertaken to verify effect of ABS in conjunction with β -TCP on healing of periodontal intrabony defect clinically and radiographically with CBCT.

SUBJECTS AND METHODS

A total of 20 intrabony periodontal defects were treated in ten patients (7 males and 3 females with an age range from 34 to 48 years). The patients were selected based on inclusion and exclusion criteria. Inclusion criteria; Presence of periodontitis with intrabony defects including premolars and molars detected by clinical and radiographic examination. (Periodontal pocket depth (PPD) \geq 6 mm and radiographic osseous defect depth \geq 4 mm), patients' ages range between (30-50) from both sexes. Exclusion criteria; Systemic diseases, radiation or immunosuppressive therapy, pregnant women or receiving contraceptive pills, smokers, patients submitted to previous periodontal surgery in the selected sites of the present study, and teeth with grade III mobility, gingival recession and furcation involvement. Patients were classified into 2 groups: 1st group (control group), ten defects were treated by β -TCP and 2nd group (test group), ten defects were treated by (mixing 0.5 ml of ABS with β -TCP).

Surgical procedure:

Before starting the surgical procedures, phase I therapy was completed for all chosen patients to provide an oral environment more favorable for wound healing

Each patient was asked to rinse his mouth with 0.2% chlorhexidine solution. All the surgical procedures were performed under local anesthesia and strict aseptic conditions. A full thickness mucoperiosteal flap was performed and complete debridement of exposed root surfaces was performed by a combination of ultrasonic and hand instrumentation.

Both ulcerated epithelium and granulation tissues on the inner surface of the flap were removed, The surgical field was irrigated with normal saline solution.

In the 1st group (control group), β -TCP bone graft was placed in the defects, while in the 2nd group (test group), 0.5 ml ABS was mixed with β -TCP bone graft then was placed in the defect. Suturing with 3-0 silk sutures were applied to

achieve primary flap closure using interrupted sutures.

Post-operative protocol:

Patients were asked to follow oral hygiene instructions and not to function over the surgical site for the initial 3 weeks. A soft diet was recommended throughout the remaining healing period. Suture removal was performed after 8-10 days.

Patients were instructed to start tooth brushing and flossing or interproximal brushing at the end of the 2nd postoperative week. Patients were followed up frequently at 6 and 12 months postoperatively.

The clinical indices measurements of Plaque index (PI)⁽¹⁰⁾, Gingival index (GI)⁽¹¹⁾, Periodontal pocket depth (PPD) and Clinical attachment level (CAL) were recorded at baseline, 6 and 12 months after surgery by the same operator. Radiographic measurements of bone density, M-D width of the defect and depth of the defect (DB) were recorded also at baseline, 6 and 12 months after surgery using CBCT.

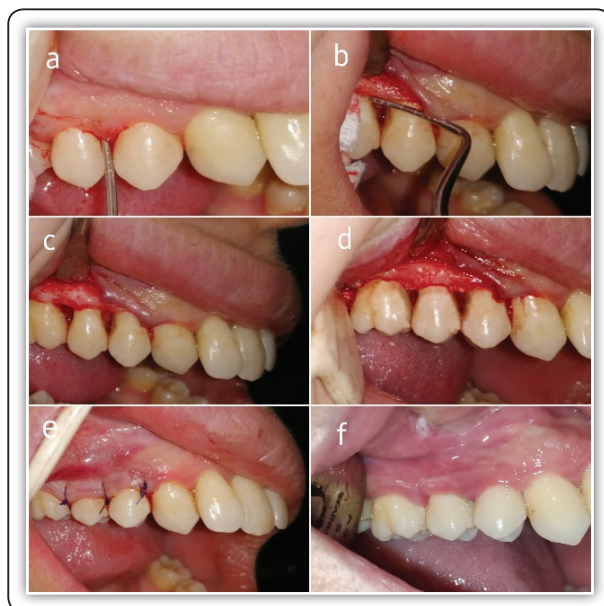


FIG (1) a; Pre-operative probing depth, b; (M-D) width of the defect, c; Flap reflection for the intrabony defect, d; Filling the intrabony defect with β -TCP mixed with ABS, e; Flap closure, f; Healing after 12 months.



FIG (2) a; Pre-operative probing depth, b; Vertical Measurement of the defect, c; (M-D) width of the defect, d; Filling the intrabony defect with β -TCP, e; Flap closure, f; Healing after 12 months.

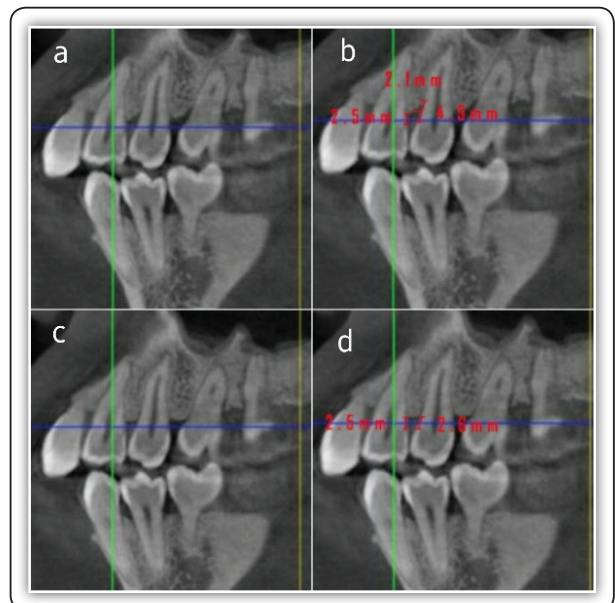


FIG (3) Corrected Sagittal view a; The intrabony defects. b; Vertical measurements of the defect. c; Bone level at 12 months. d; Vertical measurements at 12 months.

RESULTS

Table (1) showed comparison between the two studied groups according to the assessed clinical parameters. Plaque index, at baseline showed a non-statistically significant difference in the two groups. At 6 months: there was a statistically non-significant difference in mean plaque index in the two groups. At 12 months: there was a statistically significant difference ($p \leq 0.05$) in mean plaque index in the two groups. TCP/ABS group showed a less mean plaque index than TCP group.

TABLE (1): Comparison between the two studied groups according to Plaque index, Gingival index, probing pocket depth and clinical attachment loss.

	β -TCP (n =10)		β -TCP/ABS (n =10)		T	P
	Mean	\pm SD	Mean	\pm SD		
Plaque index						
At baseline	1.0	0.0	1.0	0.0	-	-
6 months	0.64	0.13	0.54	0.17	1.299	0.218
12 months	0.50	0.0	0.32	0.12	3.873*	0.008*
Gingival index						
At baseline	1.0	0.11	1.0	0.110	1.245	0.260
6 months	0.68	0.12	0.50	0.0	3.873*	0.008*
12 months	0.54	0.22	0.25	0.0	3.361*	0.015*
Probing pocket depth						
At baseline	6.11	0.09	6.24	0.13	1.183	0.149
6 months	4.43	0.13	4.31	0.20	1.265	0.230
12 months	4.53	0.13	4.36	0.24	1.654	0.124
Clinical attachment loss						
At baseline	4.19	0.12	4.21	0.12	0.440	0.668
6 months	2.44	0.10	2.33	0.13	1.903	0.081
12 months	2.56	0.08	2.33	0.13	4.086*	0.002*

t: Student t-test

p: p value for comparing between the studied groups

*: Statistically significant at $p \leq 0.05$

Concerning Gingival index, at baseline, there was a non-statistically significant difference in both groups. At 6 and 12 months, there was a statistically significant difference in mean gingival index in the two groups. TCP/ABS group showed a less mean gingival index than TCP group.

According to probing pocket depth, at baseline as well as 6 and 12 months, there was a non-statistically significant difference in mean probing pocket depth in the two groups. TCP/ABS group showed a less mean probing pocket depth than TCP group.

Clinical attachment loss, at baseline as well as 6 months revealed a non-statistically in the two groups. At 12 months, there was a statistically significant difference ($p \leq 0.05$) in both groups. TCP/ABS group showed a less mean clinical attachment loss than TCP group.

Table (2) shows comparison between the two studied groups according to M-D width of the defect ,depth of the defect and bone density. At baseline as well as 6 and 12 months, there was a non-statistically significant difference in mean M-D width of the defect in the two groups. TCP/ABS group showed a less mean M-D width of the defect than TCP group. Regarding mean depths of the defects, at baseline, there was a non-statistically significant difference in in the two groups, while at 6 and 12 months there was a statistically significant difference ($p \leq 0.05$) in both groups. TCP/ABS group showed a less mean depth of the defect than TCP group. Concerning bone density, at baseline as well as 6 months, there was a non-statistically significant difference in the two groups. At 12 months, there was a statistically significant difference ($p \leq 0.05$) in mean bone density in the two groups. TCP/ABS group showed higher mean bone density than TCP group.

TABLE (2): Comparison between the two studied groups according to M-D width of the defect, Depth of the defect and Bone density.

	β-TCP (n =10)		β-TCP/ABS (n =10)		t	P
	Mean	±SD	Mean	±SD		
M-D width of the defect						
At baseline	2.03	0.13	2.14	0.14	1.611	0.133
6 months	0.34	0.08	0.34	0.11	0.0	1.000
12 months	0.47	0.05	0.39	0.15	1.470	0.167
Depth of the defect						
At baseline	4.23	0.13	4.19	0.12	0.0	1.000
6 months	2.43	0.10	2.26	0.13	2.855*	0.014*
12 months	2.56	0.08	2.26	0.13	5.306*	<0.001*
Bone density						
At baseline	745.1	133.9	789.4	117.9	0.657	0.524
6 months	452.3	133.9	576.7	147.3	1.653	0.124
12 months	424.1	139.8	709.7	204.3	3.052*	0.010*

t: Student t-test

p: p value for comparing between the studied groups

*: Statistically significant at $p \leq 0.05$

DISCUSSION

Periodontal diseases are multifactorial infections leading to destruction of the periodontal structures, including the tooth supporting tissues, periodontal ligament, and alveolar bone (12).

CBCT used for diagnostic and quantitative measurements of alveolar bone levels in three dimensions, imaging of periodontal intrabony defects, dehiscence and fenestration defects, diagnosis of furcation-involved molars, and implant site imaging (9).

The use of ABS may have potential to enhance a positive effect on new bone formation in the wound healing process (13). Many graft materials were used in order to improve the bone tissue with various losses of material due to different reasons (14). The duration and rate of resorption are important in

bone graft materials. Early resorption may result in failure to achieve the desired bone regeneration in bone tissue (15).

β-TCP is a type of graft with late resorption features that preserves its physical structure for a long time in the defect area. Sometimes this resorption period lasts for 1–1.5 years (15).

The present split mouth study design on ten patients ranged in age between 34 – 48 years with a mean age of 40.71 ± 4.89 years (7 males and 3 females) revealed an excellent outcome in healing especially with the addition of ABS to β-TCP in the treatment of periodontal intrabony defects.

In the present study. β-TCP/ABS group showed a less mean plaque index, gingival index, clinical attachment loss, M-D width of the defect and depth of the defect than β-TCP group. Also, β-TCP/ABS group showed higher mean bone density than β-TCP group.

Overall, our data suggest that the individuals had good standards of oral hygiene, with minimal inflammatory signs around teeth.

A major limitation of our study is the lack of histological analysis to describe the characteristics of the tissues in the treated areas. Moreover, the amount of bone regeneration found in this study was based on clinical and radiographic examination.

As a result of the present study addition of ABS to Beta tricalcium phosphate in the intrabony periodontal defect showed minimal or no significant difference in clinical parameters compared to Beta tricalcium phosphate alone.

One possible factor which could explain this result can be attributed to that, ABS hasn't affected on soft tissue parameters This result are consistent with Pamuk, et al. in 2016. who explained the clinical efficacy of (ABS) when used in combination with autogenous cortical bone graft (ACB) in the treatment of intrabony periodontal defects. The authors stated that no significant differences in PI, GI, PPD observed between the groups (16).

GÜL, et al. explained effect of ABS on bone healing. The authors stated that bone promoting effect of ABS seems much complicated since ABS is an agent with anti-inflammatory, anti-thrombin, and anti-platelet effects, and it inhibits angiogenesis and neovascularization which are required for bone formation. It appears interesting when we consider that local hemorrhage seems favorable for healing process in the classical bone repair as the blood clot forms a scaffold for the osteoblasts and fibroblasts involved in bone repair, and on the other hand ABS facilitates osteogenesis while preventing hemorrhage. It may be suggested that protein network developed between ABS and blood proteins may induce other possible molecular pathways or provide a background for mesenchymal stem cells to proliferate or intervene in other unknown mechanisms crucial for bone formation⁽¹⁷⁾.

Also, İşler et al. investigated the effects of ABS on early bone healing histopathologically using a rat tibia defect model. They observed that the application of ABS decreased the occurrence of inflammation and necrosis, while increasing new bone formation in bone healing period⁽⁷⁾.

Based on the results of the present study, it could be concluded that, both treatment modalities demonstrated minimal inflammatory signs around teeth. Moreover, Ankaferd Blood Stopper® combined with β tricalcium phosphate may enhance the healing and regenerative processes and lead to more bone fill, as well as density, compared to β tricalcium phosphate alone.

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