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Original Article

Melatonin gel as an adjunct to xenograft in the surgical management of intrabony periodontal defects: A randomized controlled clinical study

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Abstract

Background. Periodontitis often leads to alveolar bone resorption, compromising tooth stability and long-term prognosis. Recently, melatonin has attracted attention for its anti-inflammatory, antioxidant, and potential osteogenic effects; however, evidence for its effectiveness in periodontal therapy remains limited. This clinical study aims to investigate the therapeutic efficacy of melatonin in periodontal surgical treatment by evaluating changes in radiographic and clinical parameters.

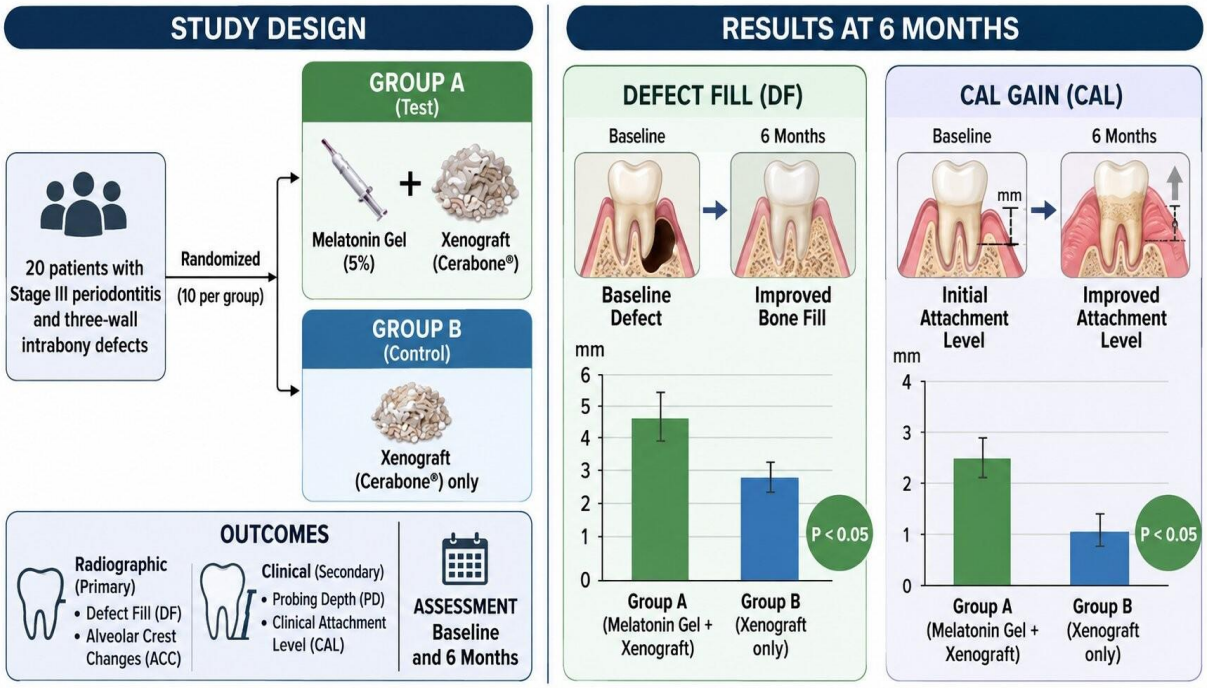
Methods. Twenty patients diagnosed with stage III periodontitis and presenting with three-wall intrabony defects were randomly assigned to either group A (melatonin gel + xenograft) or group B (xenograft only) (10 per group). The primary outcome was radiographic evaluation, including defect fill (DF) and alveolar crest changes (ACC). The secondary outcome was evaluation of clinical parameters, including probing depth (PD) and clinical attachment level (CAL). All parameters were assessed at baseline and after 6 months.

Results. At 6 months, both groups improved; however, group A showed significantly greater defect fill (4.38±0.59 mm vs. 2.88±0.56 mm; $P<0.05$) and CAL gain (2.33±0.71 mm vs. 1.11±0.33 mm; $P<0.05$) than group B.

Conclusion. Adjunctive use of melatonin gel with xenograft significantly improved bone fill and clinical attachment level compared to xenograft alone after 6 months.

The current trial was approved by Clinical Trials (Registration Number: NCT06821425).

Graphical Abstract



Key words: Clinical attachment level, intrabony defect, melatonin, periodontitis, xenograft.

Introduction

Periodontitis is a chronic, multifactorial inflammatory disease, marked by the gradual breakdown of the supporting tissues of the dentition, resulting from an exaggerated host immune-inflammatory response triggered by microbial dysbiosis.[1] It is regarded as a significant public health concern because of its widespread occurrence and its capacity to cause tooth loss, compromise oral function and aesthetics, raise healthcare expenses, and adversely impact overall well-being and quality of life.[2]

Periodontitis often leads to alveolar bone defects, which, if left untreated, may compromise the long-term survival of the involved teeth.[3,4] Although non-surgical periodontal therapy can control infection and inflammation, it is often insufficient for the management of intrabony defects, where surgical intervention is recommended.[5]

Regenerative periodontal approaches were introduced with barrier membranes, which preserve space and may facilitate the formation of new periodontal and alveolar bone tissues.[6,7] However, membrane application is often associated with complications such as higher costs, risk of exposure, and prolonged surgical time. Consequently, bone grafting without membranes has been suggested as a reliable alternative, particularly in contained three-wall intrabony periodontal defects.[8]

Bone grafts can effectively prevent epithelial cell migration into the defect, eliminating the need for a membrane, particularly in contained three-wall intrabony defects.[9] Various bone graft materials have been developed for periodontal therapy; among these, xenografts have demonstrated clinical effectiveness in managing intrabony defects.[10,11]

Xenografts, derived from bovine, equine, or porcine sources, promote bone formation through their osteoconductive properties. They offer advantages such as abundant availability, slow resorption, lower processing costs, no donor-site morbidity, and faster healing times.[12,13]

Deproteinized bovine bone has shown efficacy in radiographic bone fill both with and without barrier membranes, particularly in contained 3-wall intrabony defects.[14] Furthermore, combining xenografts with other biomaterials has been found to enhance their osteoinductive potential in addition to their inherent osteoconductive properties.[15]

Melatonin (N-acetyl-5-methoxytryptamine), primarily recognized for regulating circadian rhythms (sleep–wake cycles), is chiefly produced by the pineal gland, with additional secretion occurring in the retina, bone marrow, and gastrointestinal tract. Although once considered a hormone, it is now classified as a growth factor because it is produced in multiple tissues and has a nonspecific range of actions.[16, 17]

The therapeutic effect of melatonin is linked to its anti-inflammatory, antioxidant, antibacterial, and immunomodulatory properties.[18,19] It helps alleviate oxidative stress by directly scavenging exogenous and endogenous reactive oxygen species (ROS) as well as stimulating the function of antioxidant enzymes such as superoxide dismutase, glutathione reductase, glutathione peroxidase, and vitamin C.[20] Additionally, melatonin suppresses the release of pro-inflammatory mediators, including tumor necrosis factor-α (TNF-α), interleukin-1β (IL-1β), and interleukin-6 (IL-6).[21]

Of special relevance, melatonin was found to promote the synthesis of type I collagen fibers and influence bone formation by stimulating osteoblastic proliferation, differentiation, activation, and bone deposition while inhibiting bone loss through downregulation of the receptor activator of nuclear κ-B ligand (RANKL).[22]

Several studies have evaluated the effectiveness of melatonin in combination with non-surgical periodontal therapy (NSPT) and reported improved clinical outcomes. However, to the best of our knowledge, studies investigating melatonin in surgical settings have been limited to experimental or animal models, and no human studies have evaluated the effect of locally applied melatonin gel in the surgical management of intrabony periodontal defects.

In view of melatonin's antioxidant, host modulatory, and osteogenic actions, this study was conducted to determine the adjunctive role of melatonin gel with xenograft in surgically managing intrabony periodontal defects. The primary outcome was to evaluate changes in radiographic parameters, while secondary outcomes included changes in clinical parameters. We hypothesized that adjunctive melatonin gel would yield superior radiographic and clinical outcomes compared with xenograft alone.

Methods

Study Design and Registration

This randomized, controlled, double-blind (patient- and statistician-blinded) clinical trial with a 1:1 allocation ratio was conducted at the Department of Oral Medicine and Periodontology, Faculty of Dentistry, Ain Shams University (Cairo, Egypt), between November 2024 and July 2025. The study was conducted in accordance with the Declaration of Helsinki and approved by the Research Ethics Committee of the Faculty of Dentistry, Ain Shams University (FDASU-RecIM122330, 19 January 2024). The current trial was also approved by Clinical Trials (Registration Number: NCT06821425). Written informed consent was obtained from all participants after a full explanation of the procedure.

Sample Size Calculation

A power analysis was conducted to ensure adequate power for a two-sided statistical test of the null hypothesis that no difference exists between the tested groups in clinical assessment. By adopting an α level of 0.05, a β level of 0.2 (i.e., power=80%), and an effect size (d) of 1.599;[23] the minimum sample size required was estimated to be 16 patients (n=8 per group). Allowing for a 25% potential dropout, the total sample was increased to 20

patients (n=10 per group). A sample size calculation was performed using R statistical analysis, software version 4.3.2 for Windows.

Inclusion Criteria

The study included participants of both genders aged 35–55 years with stage III periodontitis.[24] Eligible sites exhibited residual probing pocket depth ≥6 mm, clinical attachment loss (CAL) ≥5 mm, and the presence of a three-wall intrabony defect, all evaluated six weeks after completion of initial conventional periodontal therapy.[25] Participants were required to be systemically healthy,[26] able to attend treatment sessions, comply with follow-up visits, and maintain proper oral hygiene.

Exclusion Criteria

Smokers,[27] drug abusers, pregnant or breastfeeding women,[28] members of vulnerable populations, and those with known allergies or hypersensitivity to melatonin or any component of the topical gel were excluded from the study. Individuals who received any systemic or topical medications known to interfere with periodontal healing within the last 6 months before the study were also excluded.

Patient Grouping and Randomization

Twenty participants who met the eligibility criteria were randomly distributed in a 1:1 ratio using a predetermined computer-generated sequence (www.randomizer.org). Allocation was carried out by an independent individual, not involved as the principal investigator, dividing patients into two equal groups:

- Group A (test group): Ten patients underwent open flap debridement, followed by the application of particulate xenograft mixed with 5% melatonin oral gel.
- Group B (control group): Ten patients underwent open flap debridement with particulate xenograft alone.

Sequentially numbered opaque envelopes, sealed to maintain confidentiality, were employed to conceal group assignments.

Preoperative Considerations and Evaluation

A comprehensive medical and dental history was obtained for all eligible patients according to the inclusion criteria. Before recruitment, all participants underwent professional mechanical plaque removal (PMPR), including full-mouth supra- and subgingival debridement and root surface debridement, performed with an ultrasonic scaler complemented by manual instrumentation under local anesthesia to minimize discomfort. Following the procedure, patients received instructions on self-performed biofilm control, including proper tooth brushing and rinsing twice daily for 10 days using 0.12% chlorhexidine gluconate mouthwash. The patients were instructed to avoid dental floss or interdental brushes in the treated proximal area to prevent trauma during the study period.

The periodontal reassessment was performed after six weeks to assess sites with persistent pocket depths ≥6 mm and CAL ≥5 mm, which required surgical intervention and were included in the study. At this point, a CBCT scan was performed to confirm the morphology of the three-wall intrabony defect.

Radiographic Assessment

Radiographic assessment of the defect was performed using CBCT (iCAT Next Generation Cone Beam 3D System, USA) at baseline and six months postoperatively. Imaging settings were as follows: a field of view (FOV) of 16×6 cm, a voxel size of 0.2×3 mm, and an exposure time of 6 seconds. Exposure was performed at 5 milliampere-seconds

(mAs) and 85 kVp. Standardization during imaging was established by adjusting the patient positioning lights as follows: The upper and lower light beams were adjusted to define the top and bottom of the field of view (FOV). The sagittal and lateral lights were centered to align the region of interest (ROI) within the FOV. Patients were instructed to remain motionless during exposure. The superimposition process was carried out by combining the baseline image with the image taken six months after the surgery.[29] The superimposition was achieved using Romexis 3D imaging software (Planmeca Romexis Dental software, Helsinki, Finland). The superimposition process and radiographic measurements were performed by an independent experienced radiologist. The radiologist was blinded to the clinical outcomes and time points to minimize potential bias.

The following views were evaluated on CBCT:

Baseline CBCT: The axial and sagittal views were evaluated to assess the morphology of the three-wall intrabony defect to be included in the study.

Baseline and postoperative CBCT: The sagittal views were evaluated to measure the distance from the CEJ to the base of the defect (CEJ-BD) (Figure 1a, b) as well as the distance from the CEJ to the alveolar bone crest (CEJ-AC).

Clinical Assessment

Clinical parameters at the target sites were assessed using a UNC-15 periodontal probe (Hu-Friedy®, Chicago, USA) six weeks after conventional therapy (baseline) and again six months after surgery. Pocket depth (PD)[30] was measured from the gingival margin to the pocket base, while clinical attachment level (CAL)[30] was calculated from the CEJ to the pocket base. The same operator recorded all clinical measurements using standardized probing force and measurement protocols to ensure consistency and reduce operator-related bias.

Histological Assessment

Histological evaluation was not feasible because of ethical constraints on obtaining tissue biopsies from periodontal defects in human subjects. Therefore, clinical and radiographic outcomes were used as objective measures of healing and defect fill.

Melatonin Oral Gel Preparation

A 5% melatonin oral gel[31] was formulated by Nanogate Scientific Lab (Cairo, Egypt) using hydroxypropyl methylcellulose (HPMC, 1.5% w/v, high-viscosity grade) as the gelling agent. HPMC was dispersed in distilled water at 80°C under continuous stirring, after which melatonin was incorporated to complete the final volume. The gel was degassed under vacuum, adjusted to pH 6.8 for optimal stability and tissue compatibility, and stored at 4°C until use. The formulation was prepared under aseptic conditions, with the melatonin solution sterilized by 0.00022 mm filtration and all components handled in a laminar-flow cabinet. The present study did not evaluate the release profile of melatonin from the HPMC-based gel.

Particulate Xenograft

Bovine particulate xenograft (0.5–1.0-mm particles; Botiss Cerabone, Germany) was used to fill the intrabony defects in both groups.

Surgical Procedures Performed

Surgical sites were anesthetized with 4% articaine and 1:200,000 epinephrine (Artinibsa, Spain), followed by a sulcular incision using a 15c sterile carbon steel blade (Tekno, Germany). Full-thickness buccal and lingual/palatal flaps were reflected, with care taken to preserve the interdental papillae, achieve adequate flap reflection, and avoid trauma

to adjacent teeth and soft tissues. Granulation tissue was meticulously removed, and the root surfaces were thoroughly debrided in both groups, followed by irrigation with sterile saline (Figure 2). Strict aseptic technique was followed throughout the procedure. The same periodontist performed all surgeries using standardized surgical protocols to ensure consistency and reduce operator-related variability.

- Group A (test group): The intrabony defect was grafted using 0.25 mL of particulate xenograft blended with 0.25 mL of melatonin gel, flushing with the alveolar crest (Figure 3).
- Group B (control group): The intrabony defect was grafted with particulate xenograft alone, with the alveolar crest also flushed.

The flaps were subsequently placed back and secured using a simple interrupted technique with 5-0 polypropylene sutures (D-tek polypropylene suture, Cyprus).

Postoperative Recommendations and Considerations

Postoperative care included rinsing with 0.12% chlorhexidine gluconate for 14 days. Sutures were removed one week after surgery, and patients were recalled monthly for professional prophylaxis and oral hygiene maintenance. Patients were advised to avoid trauma or pressure on the surgical site and to maintain careful oral hygiene.

Results

Statistical Analysis

Categorical data were expressed as frequencies (n) and percentages (%) and analyzed using Fisher's exact test. Numerical data were presented as mean ± standard deviation (SD). Data distribution was assessed by visual inspection and the Shapiro–Wilk test. Radiographic measurements, probing depth (PD), and clinical attachment level (CAL) were normally distributed and analyzed using two-way mixed-effects linear models. Model assumptions were assessed by evaluating the normality of residuals, linearity, and homoscedasticity, with the latter assessed using the Breusch–Pagan test. Age was normally distributed and compared between groups using an independent-samples t-test. Percentage changes in clinical and radiographic parameters were non-normally distributed and were therefore compared between groups using the Mann–Whitney U test. Statistical significance was set at $P<0.05$. All statistical analyses were performed using R statistical analysis software, version 4.5.0, for Windows (R Core Team, 2025).

Twenty patients were recruited; two patients did not complete the study and dropped out. Thus, statistical analyses were performed on 18 patients (i.e., 9 cases per group). The test group consisted of 2 males (22.22%) and 7 females (77.78%), whereas the control group included 1 male (11.11%) and 8 females (88.89%). The mean (SD) age of the cases in the test group was 40.44 years, while it was 39.11 years in the control group. No statistically significant differences were found between the groups in terms of gender ($P=1$) or age ($P=0.567$). Furthermore, no serious side effects were observed in patients treated with 5% melatonin oral gel. Four patients (n=2 per group) experienced mild postoperative edema, which resolved spontaneously without any intervention.

Regarding the primary outcome, radiographic measurements, including the distance from the CEJ to the base of the defect (CEJ–BD) and the distance from the CEJ to the alveolar crest (CEJ–AC) at baseline and 6 months post-treatment, Intragroup analysis revealed a significant reduction in both parameters from baseline to 6 months. In terms of absolute changes, the melatonin group showed greater defect fill (DF) (4.38±0.59 mm) compared with the control group (2.88±0.56 mm). Moreover, intergroup comparison of percentage changes demonstrated that the melatonin group exhibited significantly greater

improvements in CEJ–BD and CEJ–AC compared with the control group (CEJ–BD: 54.80±5.31% vs. 37.28±4.86%; P<0.001; CEJ–AC: 34.54±5.47% vs. 20.42 ± 9.13%; P=0.002), as shown in Tables 1 and 2 and Figures 4 and 5.

Regarding the secondary outcome, clinical parameters, including probing depth (PD) and clinical attachment level (CAL), intragroup analysis showed a significant reduction in both measures after 6 months. In absolute terms, the melatonin group showed greater CAL gain than the control group (2.33±0.71 mm vs. 1.11±0.33 mm). Intergroup comparison of percentage changes further indicated that the test group achieved significantly greater improvements in PD and CAL than the control group (PD: 62.83±5.82% vs. 52.01±10.05%; P=0.016; CAL: 56.11±11.96% vs. 30.56±8.33%; P=0.001) (Tables 3 and 4 and Figures 6 and 7).

Discussion

Melatonin is a multifunctional pleiotropic molecule with diverse roles, including immunomodulatory, anti-inflammatory, and antioxidant activities.[32] The potential mechanisms underlying the observed clinical and radiographic improvements may be related to biological effects of melatonin reported in previous studies, including effects on osteoblast proliferation and differentiation, VEGF-related angiogenesis, and RANKL/OPG modulation. Nevertheless, these mechanisms remain hypothetical in the context of the present study, as no biomarker, cellular, or molecular assessments were performed.[33,34] Therefore, the hypothesis of this trial was that using melatonin gel in conjunction with xenograft in surgically managing intrabony periodontal defects would yield better outcomes compared to using xenograft alone.

Bovine-derived particulate xenograft was applied in both groups because of its osteoconductive property, biocompatibility, and slow rate of resorption.[12,13] Thus, it prevents soft tissue collapse within the intrabony defect, giving enough time for the bone fill to occur. Hydroxypropyl methylcellulose was selected as a melatonin carrier because of its hydrophilicity, bioadhesiveness, surface properties for cell adhesion, controlled-release matrix, cell viability, non-toxicity, excellent biocompatibility, and biodegradability.[35] However, no formal assessment of the drug release profile or long-term stability of the formulation was conducted, which represents a limitation of the present study.

The radiographic changes were analyzed using CBCT, which enables accurate, detailed images of intrabony defects, including mesiodistal and faciolingual measurements, without distortion from overlapping anatomical structures.[36] It also proves to be a reliable tool in monitoring the outcomes of periodontal surgical treatments.[37] A UNC graduated probe was employed to assess the clinical outcomes. Thus, combining radiographic findings with clinical parameters may provide a more comprehensive assessment of changes in radiographic defect fill and clinical periodontal outcomes following periodontal surgical treatment.

No significant differences were detected between the two groups regarding dental arch (P=1) or tooth distribution (P=1). Baseline radiographic and clinical parameters also did not differ significantly between groups, ensuring that treatment results could be reliably compared throughout the follow-up periods.

At six months, intragroup analysis of CEJ–BD showed a highly significant reduction in both the test and control groups (P<0.001 for each), confirming considerable defect resolution in both interventions. This may be explained by the favorable morphology of three-wall intrabony defects, which provide a spatial configuration for blood clot stabilization.[38] Moreover, application of xenograft following thorough debridement of three-wall intrabony periodontal defects can result in substantial bone fill.[14]

When comparing groups, the test group exhibited a significantly greater percentage reduction in CEJ–BD than the control (54.80±5.31% vs. 37.28±4.86%, P<0.001), indicating more defect fill with melatonin. This effect may be attributed to its role in inhibiting NF-κβ and RANKL, which are involved in impaired bone formation and osteoblast inhibition.[39,40] These findings align with a previous study,[41] which showed that bony defects treated with NSPT + 1% melatonin gel had significantly greater hard tissue fill (1.46±0.58 mm) compared to those treated with placebo gel (0.50±0.38 mm, P=0.0001). Similarly, periodontitis patients treated with 1% melatonin gel and NSPT had more defect fill and higher alveolar bone density than the control group (P<0.001 and P=0.028, respectively).[42] More recently, melatonin-loaded nanoparticles as adjunctive therapy to NSPT showed the greatest improvement in bone defect parameters at 6 months (P<0.05).[43]

Regarding alveolar crest changes (ACC), both groups demonstrated a gain in alveolar crest height, despite crestal bone resorption typically occurring after flap procedures.[44] However, previous studies have suggested that changes following treatment of intrabony defects may occur predominantly within the intrabony component, with minimal or no crestal resorption.[45] In the present trial, the melatonin group exhibited a significantly greater percentage gain in CEJ–AC compared with controls (34.54±5.47% vs. 20.42±9.13%, P=0.002), indicating greater alveolar crest height gain with melatonin.

Both groups achieved significant reductions in probing depth (PD) and improvements in clinical attachment level (CAL) at six months, most likely due to thorough debridement and xenograft use. Likewise, a more recent review by Stavropoulos et al.[46] found that regenerative techniques were associated with greater PD reduction and CAL gain than OFD alone.

Between groups, the test group showed significantly greater percentage improvements in PD and CAL than controls (P=0.016 for PD; P=0.001 for CAL). These outcomes align with multiple clinical studies[47-51] that reported enhanced PD reduction and CAL gain with melatonin used either topically or systemically as an adjunct to NSPT. The beneficial effects are linked to melatonin's antioxidant, anti-inflammatory, immunomodulatory, and antibacterial properties.[52] Similar improvements were noted in gingival index and pocket depth following topical melatonin use.[32]

Limitations of this study include the lack of assessment of the melatonin release profile, which restricts the ability to determine the kinetics of gel delivery and its sustained availability at the surgical site. The relatively short follow-up period limits conclusions regarding the long-term stability of clinical improvements. The absence of a radiographic stent may have introduced minor variability in defect measurements, despite the superimposition technique used to improve standardization and comparability of serial radiographic measurements by aligning baseline and follow-up scans using stable anatomical structures. Ethical considerations prevented histological evaluation, which would have allowed direct confirmation of true periodontal regeneration. The potential operator bias resulting from the lack of blinding should be considered when interpreting the results. The relatively small final sample size represents an important limitation of the present study. Although an a priori sample size calculation was performed and the initial recruitment target was increased to account for potential dropouts, the final analysis included only 18 participants (9 per group). Therefore, the findings should be interpreted cautiously, and randomized controlled trials with larger cohorts, standardized radiographic protocols, and longer follow-up periods are warranted to confirm the present findings.

Conclusion

Within the trial's limitations, it may be concluded that the adjunctive use of melatonin gel with xenograft produced significantly greater radiographic and clinical improvements than xenograft alone at 6 months.

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Competing Interests
The authors declare no conflicts of interest.

Data Availability
Data used in this study are available from the corresponding author upon request.

Ethical Approval
This study was conducted in accordance with the Declaration of Helsinki and was approved by the Research Ethics Committee of the Faculty of Dentistry, Ain Shams University (Approval No. FDASU-RecIM122330).

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Table 1. Summary statistics and comparative analysis of CEJ-BD (mm) measurements between both groups, including intragroup changes

Time	CEJ-BD (mm) (Mean ± SD)		P-value (groups)	Effect size (95% CI)**
	Test (n=9)	Control (n=9)		
Baseline	8.04±1.23	7.68±0.90	0.418	0.00 (0.00 to 0.00)
6 months	3.66±0.82	4.80±0.54	0.015*	0.22 (0.00 to 0.50)
P-value (intervals)	<0.001*	<0.001*		
Reduction (mm)	4.38±0.59	2.88±0.56	<0.001*	1.00 (1.00 to 1.00)
Percentage of change	54.80±5.31	37.28±4.86	<0.001*	1.00 (1.00 to 1.00)

**Omega squared for individual measurements and rank biserial correlation coefficient reduction and percentage change data; *: Significant at P<0.05

Table 2. Summary statistics and comparative analysis of CEJ-AC (mm) measurements between both groups, including intragroup changes

Time	CEJ-AC (mm) (Mean ± SD)		P-value (groups)	Effect size (95% CI)**
	Test (n=9)	Control (n=9)		
Baseline	4.50±1.11	3.97±0.66	0.205	0.04 (0.00 to 0.31)
6 months	2.98±0.91	3.16±0.65	0.653	0.00 (0.00 to 0.00)
P-value (intervals)	<0.001*	<0.001*		

Reduction (mm)	1.52±0.29	0.81±0.36	0.002*	0.89 (0.70 to 0.96)
Percentage of change	34.54±5.47	20.42±9.13	0.002*	0.88 (0.67 to 0.96)

**Omega squared for individual measurements and rank biserial correlation coefficient reduction and percentage change data; *: Significant at P<0.05

Table 3. Summary statistics and comparative analysis of PD (mm) measurements between both groups, including intragroup changes

Time	**PD (mm) (Mean ± SD)**		**P-value (groups)**	**Effect size (95% CI)****
	Test (n=9)	**Control (n=9)**		
Baseline	6.89±1.17	6.78±1.09	0.797	0.00 (0.00 to 0.00)
6 months	2.56±0.53	3.22±0.67	0.130	0.05 (0.00 to 0.27)
P-value (intervals)	<0.001*	<0.001*		
Reduction (mm)	4.33±0.87	3.56±1.01	0.116	0.43 (-0.09 to 0.77)
Percentage of change	62.83±5.82	52.01±10.05	0.016*	0.67 (0.25 to 0.87)

**Omega squared for individual measurements and rank biserial correlation coefficient reduction and percentage change data; *: Significant at P<0.05

Table 4. Summary statistics and comparative analysis of CAL (mm) measurements between both groups, including intragroup changes

Time	**CAL (mm) (Mean ± SD)**		**P-value (groups)**	**Effect size (95% CI)****
	Test (n=9)	**Control (n=9)**		
Baseline	4.11±0.78	3.67±0.50	0.115	0.06 (0.00 to 0.30)
6 months	1.78±0.44	2.56 ± 0.53	0.009*	0.21 (0.01 to 0.47)
P-value (intervals)	<0.001*	<0.001*		
Reduction (mm)	2.33±0.71	1.11±0.33	0.002*	0.83 (0.56 to 0.94)
Percentage of change	56.11±11.96	30.56±8.33	0.001*	0.90 (0.73 to 0.97)

**Omega squared for individual measurements and rank biserial correlation coefficient reduction and percentage change data; *: Significant at P<0.05

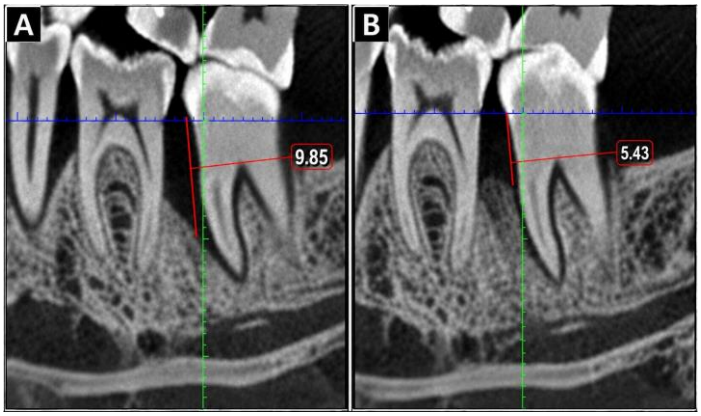

Figure 1. Representative CBCT images showing baseline defect morphology prior to surgery (a) and radiographic bone fill following regenerative periodontal therapy (b).

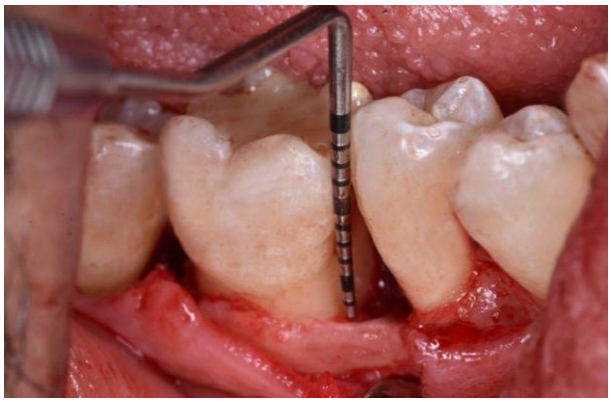

Figure 2. Intraoperative photograph demonstrating thorough mechanical debridement of the periodontal defect and root surfaces to establish a clean surgical environment for regeneration.

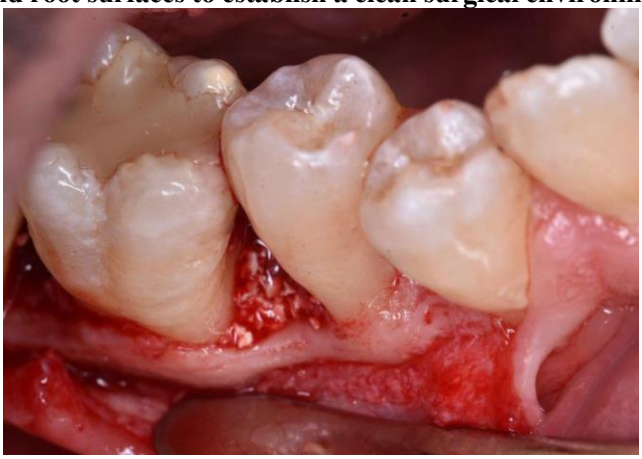

Figure 3. Intraoperative photograph demonstrating placement of the intrabony defect graft using particulate xenograft blended with melatonin gel up to the level of the alveolar crest.

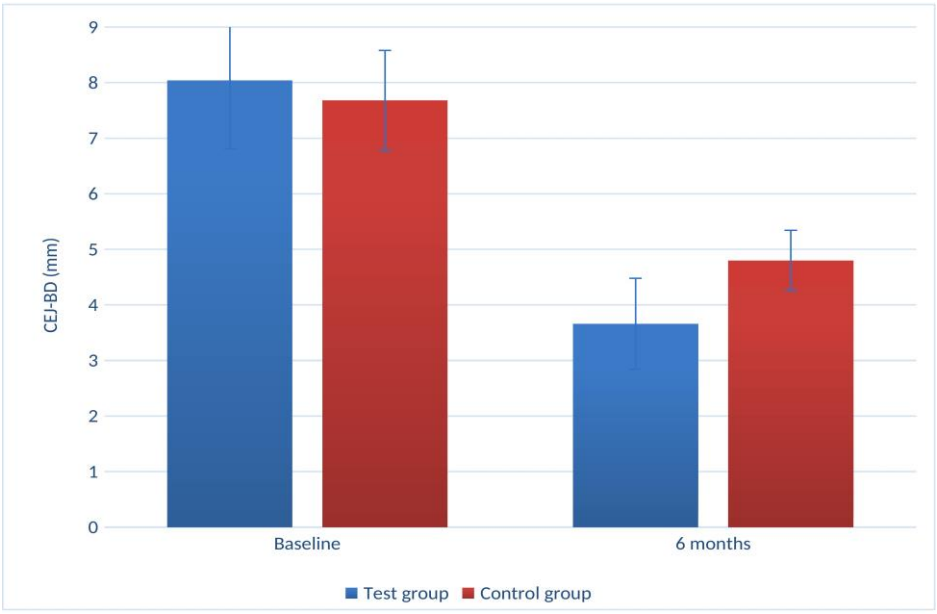

Figure 4. Bar chart comparing the absolute changes in CEJ–BD (mm) between the control and melatonin groups at 6 months postoperatively, showing significantly greater radiographic improvement and defect fill in the melatonin-treated group.

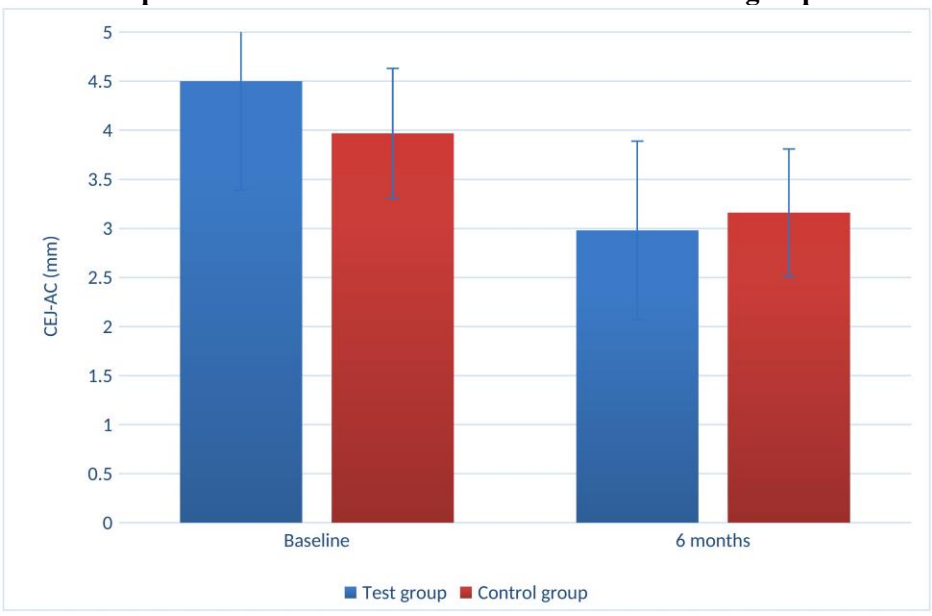

Figure 5. Bar chart comparing the absolute changes in CEJ–AC (mm) between the control and melatonin groups at 6 months postoperatively, showing greater radiographic improvement in the melatonin-treated group.

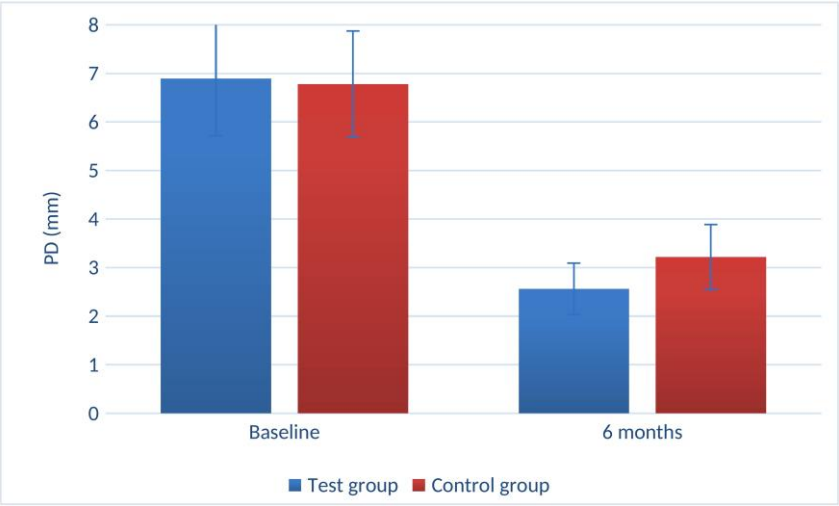

Figure 6. Bar chart comparing the absolute changes in PD (mm) between the control and melatonin groups at 6 months postoperatively, showing greater pocket depth reduction in the melatonin-treated group.

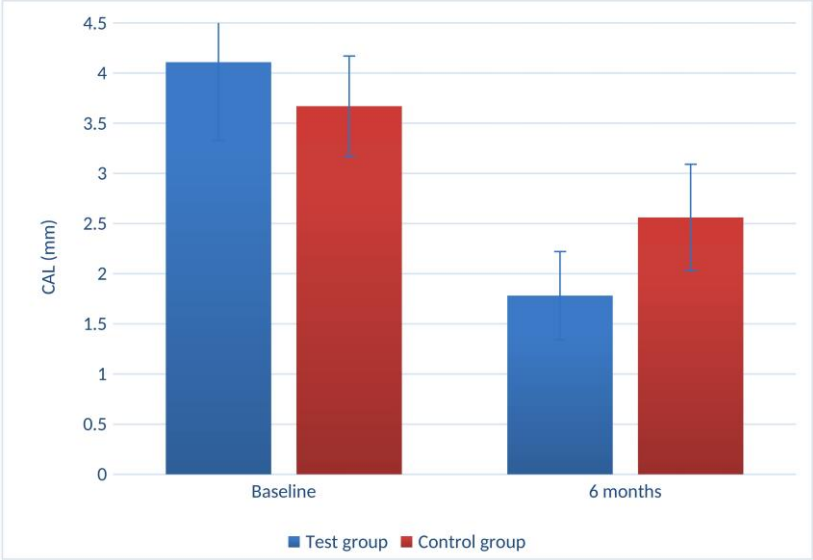

Figure 7. Bar chart comparing the absolute changes in CAL (mm) between the control and melatonin groups at 6 months postoperatively, showing significantly greater clinical attachment gain in the melatonin-treated group.