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ARTICLE

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# Synergistic Effects of Coenzyme Q10 and Calcium Carbonate Composite on Mandibular Bone Regeneration in Rabbit Model

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## ABSTRACT

**Background:** Bone healing is a complex process involving the formation of new bone to repair defects. Various supplements, such as Coenzyme Q10 and Calcium carbonate, have been studied for their potential to enhance bone regeneration. Coenzyme Q10 Act as antioxidant that reduces oxidative stress and may promote osteoblast activity, while calcium carbonate provides calcium ions necessary for bone mineralization. Combining two agents together could improve bone healing outcomes, particularly in surgical defects of the mandibular bone.

**Objectives:** The goal of this study was to investigate the effect of Coenzyme Q10 and/or calcium carbonate on blood N-Telopeptides of Type I Collagen (NTx) and Bone-specific Alkaline Phosphatase (BALP) levels, as well as the histological effects in rabbits at the healing site in the mandibular bone following surgery.

**Methods:** In a randomized controlled trial, sixty male New Zealand rabbits underwent surgical creation of a standardized mandibular bone defect. The animals were allocated to six groups (n=10/group): a negative control (empty defect) and five treatment groups where the defect was filled with either Coenzyme Q10, $CaCO_3$, or one of three $CaCO_3$: Coenzyme Q10 ratio mixtures (1:1, 1:2, 2:1). Histological and biochemical analyses were performed to assess bone healing at 15 and 30days post-operation.

**Results:** All treated groups exhibited a significant decrease in NTx and a significant increase in BALP levels compared to control values at 30 days, confirming a positive shift in bone metabolism. Histological analysis indicated enhanced bone regeneration in all treatment groups compared to the control, with the Coenzyme Q10 + $CaCO_3$ (1:2) composite showing the most substantial improvement, as evidenced by increased bone fill and osteoblast activity. A one-way ANOVA identified statistically significant differences in bone healing outcomes between the groups ($p < 0.05$).

**Conclusions:** This study found that a 2:1 calcium carbonate/Coenzyme Q10 composite graft significantly enhances mandibular bone regeneration by dual osteogenic and anti-resorptive mechanisms, supporting its potential as a smart bone graft material.

## Introduction

Bone, a dynamic mineralized tissue, possesses a remarkable capacity for regeneration. However, critical-sized defects, such as those often encountered in maxillofacial and mandibular surgery, overwhelm this innate healing potential, necessitating surgical intervention with bone graft materials [1]. Autografts remain the clinical gold standard, their use is limited by donor site morbidity and limited availability [2]. This has driven the search for innovative synthetic bone graft substitutes that can actively promote osteogenesis.

Calcium carbonate ($CaCO_3$) has emerged as a promising biomaterial due to its excellent osteoconductivity, biocompatibility, and gradual resorption profile, which is thought to be more

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synchronized with new bone formation than traditional calcium phosphates [3, 4]. Separately, coenzyme Q10, a critical component of the mitochondrial electron transport chain, has been shown to enhance cellular bioenergetics and mitigate oxidative stress, creating a microenvironment conducive to osteoblast differentiation and activity [5, 6].

Despite the individual promise of these compounds, their synergistic potential for bone regeneration remains largely unexplored. It is hypothesized that the osteoconductive scaffold provided by $CaCO_3$ could be significantly enhanced by the addition of Coenzyme Q10's bioenergetic and antioxidant properties. Therefore, this study aims to investigate the novel combination of $CaCO_3$ and coenzyme Q10 in a mandibular defect model, seeking to determine if their synergistic application results in superior bone healing compared to either compound alone.

Coenzyme Q10 is a vitamin-like substance found in most eukaryotic cells that are oily-soluble. Coenzyme Q10 is a molecular component of 1, 4-benzoquinone that has a quinone chemical group and ten isoprenyls in its tail [7].

Coenzyme Q10 is a key component of the aerobic energy generation electron transport chain (ETC), which converts ADP to ATP [8].

Coenzyme Q10 is found in the greatest amounts in organs such as the heart, liver, and kidney due to high energy needs. [9]. Because of its location in the electron transport chain (ETC) and interaction with reactive oxygen species (ROS), Coenzyme Q10 is mostly found in mitochondria [10]. Coenzyme Q10's therapeutic efficacy is critically dependent on its redox state. The reduced form, ubiquinol, demonstrates superior bioavailability and potent antioxidant capacity compared to its oxidized counterpart, ubiquinone [11]. This distinction is paramount in bone regeneration, where pathological levels of oxidative stress disrupt the delicate balance between osteoblast-mediated formation and osteoclast-driven resorption [12]. Recent studies specifically attribute ubiquinol's anti-resorptive mechanism to its ability to scavenge reactive oxygen species (ROS), thereby directly protecting osteoblasts from apoptotic signals and potently suppressing RANKL-induced osteoclastogenesis [13,14]. To leverage this targeted mechanism, this study developed a novel calcium carbonate/ubiquinol composite graft designed to actively orchestrate mandibular bone healing through concurrent osteogenic stimulation and osteoclastic inhibition.

Calcium carbonate's chemical formula is $CaCO_3$. It is the main component of eggs, snail shells, seashells, and pearls. It is a naturally occurring chemical that may be found in rocks like calcite and aragonite minerals, most notably calcite, a sedimentary rock made primarily of calcite [15]. Although it can be dangerous and cause poor digestion, it is used therapeutically as an antacid or calcium supplement [16].

As the basic module, calcium is essential for bone development and metabolism, especially for bone turnover. According to a number of studies, replacing bone with eggshells is a viable and affordable solution [17]. Numerous techniques and materials were researched in order to provide a viable substitute for autologous bone in the treatment of osseous defects. The gold standard for augmentation is the use of autologous bone, which is the recommended method [18].

## Methods

### Bone Biomarker

A biomarker is defined as a characteristic that can be objectively measured and evaluated as an indicator of normal biological processes, pathogenic processes, or pharmacological responses to a therapeutic intervention [19]. In the context of bone metabolism, specific biomarkers of bone turnover are released into the circulation during the bone remodeling cycle. These biomarkers are categorized into that reflecting bone formation

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and that reflecting bone resorption, providing a dynamic assessment of skeletal activity [20]. Bone formation and resorption occur during bone remodeling in organized cellular units known as basic multicellular units (BMUs); hence, levels of bone turnover indicators indicate formation and resorption within a BMU as well as the number of BMUs [21].

**Bone-specific Alkaline Phosphatase**

Alkaline phosphatase (ALP) is a glycoprotein found in the plasma membrane that catalyzes the release of inorganic phosphate via the hydrolysis of phosphate monoesters. These enzymes are naturally found in almost all human tissues [22]. The blood contains four ALP isoforms or isozymes: germ cells, placental, intestinal, and non-specific forms. The tissue non-specific alkaline phosphatase (TNSALP) gene generates ALP isoforms in the kidneys, liver, and bone. Around 95% of total ALP is composed of liver and bone isoforms. [23]. Bone-specific Alkaline Phosphatase is produced by osteoblasts and has a positive correlation with the pace of bone production; it is a key signaling marker for bone growth [24].

**N-Telopeptides of Type I Collagen (NTx)**

Type I collagen telopeptides are the most well-studied and used bone resorption indicators [25]. Depending on the collagen cross-linking location, two forms are released during collagen degradation: the N-terminal telopeptide (NTx) and the C-terminal telopeptide (CTx) [26]. NTx and CTx can be detected in both urine and serum [27].

**Materials and Methods**

This study was a randomized, controlled animal trial conducted over a 30-day period at the College of Dentistry, University of Mosul. The protocol received prior approval from the University of Mosul Animal Research Ethics Committee REC reference no. (UoM.Dent/A.L38/21), and all procedures adhered to institutional guidelines for the care and use of laboratory animals.

**Animals and Housing**

Sixty healthy, skeletally mature, white male New Zealand rabbits (age: 5-6 months; weight: 1.5-2.5 kg) were used. The animals were housed individually in cages under standard laboratory conditions (temperature: 23 ± 2°C; natural light/dark cycle) and had ad libitum access to a standard diet of water, maize, crushed wheat, and vegetables throughout the acclimatization and experimental periods.

**Calcium Carbonate Production from Egg Shells**

The chicken eggshell used in this study was purchased from nearby marketplaces. After removing the egg casings and removing the cover from the peel lining, the egg was carefully washed and allowed to dry. Eggshells were ground in a coffee grinder [28, 29] then purified using a particular mash (100µm) to create fine crystals. After that, 100 milliliters of distilled water were added, and the mixture was filtered through Whatman filter paper (No. 6). These produced white powder and calcium carbonate crystals after being dried and sterilized for an hour at 125 °C in an autoclave. Coenzyme Q10 was combined with calcium carbonate crystals in several weight-to-weight ratios (0:1, 1:0, 1:1, 1:2 and 2:1).

**Surgical Procedure**

General Anesthesia was induced via intramuscular injection of a ketamine hydrochloride (40 mg/kg) and xylazine (5 mg/kg) cocktail as described by Rani et al. [30]. The right mandibular region was shaved and aseptically prepared with povidone-iodine. A 5-cm longitudinal skin incision was made on the anterolateral aspect of the mandible. After blunt dissection of the soft tissues and muscle to expose the bone surface, a standardized unicortical groove defect (2 mm in diameter, 6 mm in length) was created using a bone drill under continuous saline irrigation to prevent thermal necrosis.

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**Experimental Groups and Randomization**

The animals were randomly allocated into six experimental groups (n=10/group). The defect in each group was treated as follows:

**Control Group:** The defect was left empty.

**Group I:** Defect filled with Coenzyme Q10.

**Group II:** Defect filled with pure $CaCO_3$.

**Group III:** Defect filled with a 1:1 ratio composite of $CaCO_3$ and Coenzyme Q10.

**Group IV:** Defect filled with a 1:2 ratio composite of $CaCO_3$ and Coenzyme Q10.

**Group V:** Defect filled with a 2:1 ratio composite of $CaCO_3$ and Coenzyme Q10.

**Postoperative Care and Euthanasia**

The surgical incision was closed in layers using black silk sutures and disinfected with 2% iodine. To prevent infection, including control, all animals received a postoperative intramuscular antibiotic (oxytetracycline, 0.5g) once daily for five days. Half of the animals from each group (n=5) were euthanized at two predetermined time points: 15 days and 30 days post-operation.

**Sampling of Blood**

Following animal sacrifice at the conclusion of the tests, blood samples were extracted from the jugular vein (15, 30) days. After being placed in plain tubes, five milliliters of blood were allowed to rest at room temperature for half an hour. Centrifugation was used for 10 minutes at 3000 rpm to separate the serum from the whole blood. A micropipette was used to transfer the serum to an Eppendroff tube, which was then frozen at -20 °C until it thawed and could be analyzed using a microplate ELISA reader.

**Statistics**

All quantitative data are presented as mean ± standard deviation (SD). The assumption of normality was verified using the Shapiro-Wilk test. Statistical analysis was performed using SPSS version 25.0. To determine the presence of statistically significant differences between the six groups (control and five experimental groups), a one-way analysis of variance (ANOVA) was conducted. This test compares the means of three or more independent groups to determine if there is overall evidence of a significant difference.

Following a significant ANOVA result ($p \leq 0.05$), post-hoc comparisons were carried out using Duncan's test. This test is used to identify which specific group means differ from each other by comparing all possible pairs of groups while controlling for Type I error. The criterion for statistical significance for all tests was set at a p-value of $\leq 0.05$.

**Histological Criteria**

Within 15 and 30 days after surgery, five animals from each group were euthanized in order to produce tissues for histological analysis. Biopsies were taken from the surgical site after the jaw bones were removed. Depending on the specimen thickness and degree of calcification, the specimens were decalcified in 30% formic acid for 2-4 weeks after being preserved in a 10% buffered formalin solution. Until the bone softened, the decalcification solution was replaced every 48 hours. The bone was then washed in water for a minimum of 24 hours (NBF). It then became dehydrated as the ethanol content increased. A microtome was used to cut the specimens into thin slices (5 µm thick), which were then cleaned with xylene and placed in paraffin wax. Finally, haematoxylin and eosin (H&E) staining was applied to the specimens.

Results

**1. Bone Formation Marker (BALP)**

**A. At 15 Days Post-Operation:**

**Group I** showed the lowest increase (0.69 ± 0.23 ng/ml).

**Groups II and III** demonstrated intermediate and statistically similar levels (0.86 ± 0.09 ng/ml and 0.91 ± 0.11 ng/ml, respectively).

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**Groups IV and V** displayed the highest BALP levels (1.11 ± 0.03 ng/ml and 1.15 ± 0.17 ng/ml, respectively), which were statistically equivalent to each other but significantly greater than all other groups.

Bone-specific alkaline phosphatase (BALP) activity was significantly higher in all treatment groups compared to the control group (0.27 ± 0.05 ng/ml). The treatment groups exhibited a dose- or intensity-dependent increase in BALP levels, as showed in Table 1.

**Table 1: Serum BALP levels (ng/ml) at 15 days.**

| Follow-up period 15 day BALP | |
|---|---|
| Control group | 0.27±0.05 [A] |
| Group I | 0.69±0.23 [B] |
| Group II | 0.86±0.09 [CB] |
| Group III | 0.91±0.11 [C] |
| Group IV | 1.11±0.03 [D] |
| Group V | 1.15±0.17 [D] |

Different letter mean there is significant difference, while same letter mean there in no significant difference at P ≤ 0.05.

**B. At 30 Days Post-Operation:**

Groups IV and V demonstrated the most potent effect, with the highest BALP levels (1.72 ± 0.12 ng/ml and 1.89 ± 0.22 ng/ml, respectively).

A significant, step-wise increase was observed through Group I (1.05 ± 0.21 ng/ml), Group II (1.31 ± 0.25 ng/ml), and Group III (1.48 ± 0.43 ng/ml).

The control group's BALP level was significantly lower than all treated groups except for Group I, where the difference did not reach statistical significance.

At the 30-day mark, all treatment groups showed elevated Bone-specific Alkaline Phosphatase (BALP) levels compared to the control (0.76 ± 0.19 ng/ml). The osteogenic response followed a clear gradient, as showed in Table 2.

**Table 2:** Serum BALP levels (ng/ml) at 30 days.

| Follow-up period 30 day BALP | |
|---|---|
| Control group | 0.76±0.19 [A] |
| Group I | 1.05±0.21 [BA] |
| Group II | 1.31±0.25 [CB] |
| Group III | 1.48±0.43 [DC] |
| Group IV | 1.72±0.12 [ED] |
| Group V | 1.89±0.22 [E] |

Different letter mean there is significant difference, while same letter mean there in no significant difference at P ≤ 0.05.

The BALP changed significantly between the control group, groups I, II, III, IV, and V, respectively, over the course of the 15 and 30-day study period, according to the ANOVA. Figure 1

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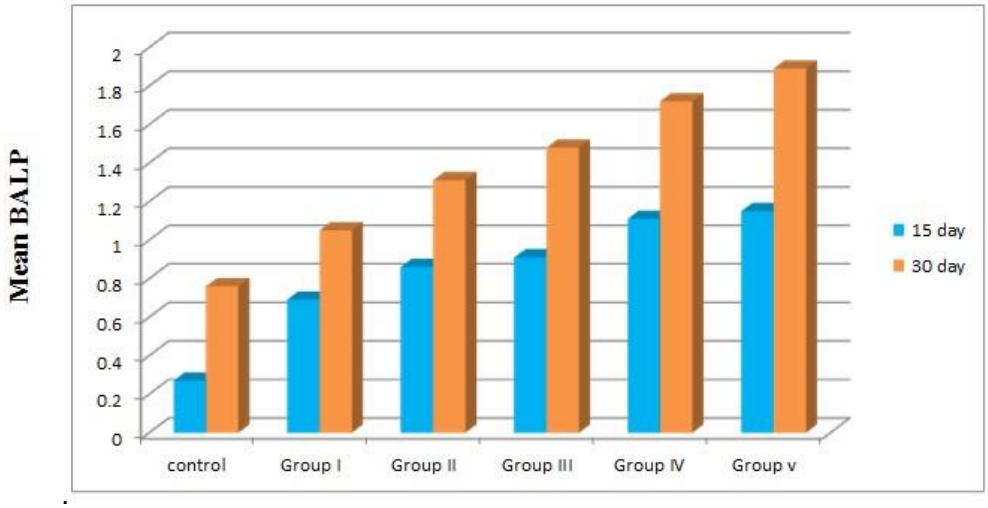


**Figure 1.** variations in BALP serum activity between the treatment and control groups throughout various follow-up times.

**2. Bone Resorption Marker (NTx)**

**A. At 15 Days Post-Operation:**

As showed in Table 3, at the 15-day mark, levels of the bone resorption marker NTx were similar across all groups, including the control. No treatment demonstrated a significant effect on bone resorptive activity at this early time point.

**Table 3:** Serum NTx levels (nmol/L) at 15 days.

| Follow-up period 15 day NTx | |
|---|---|
| Control group | 1.69±0.11 [A] |
| Group I | 1.58±0.27 [A] |
| Group II | 1.63±0.26 [A] |
| Group III | 1.48±0.31 [A] |
| Group IV | 1.64±0.09 [A] |
| Group V | 1.59±0.15 [A] |

Same letter mean there in no significant difference at $P \leq 0.05$.

**B. At 30 Days Post-Operation:**

Group V demonstrated the strongest anti-resorptive effect (1.0 ± 0.16 nmol/L).

Groups III and IV showed a strong and statistically similar level of suppression (1.83 ± 0.33 nmol/L and 1.7 ± 0.22 nmol/L, respectively).

Groups I and II provided a more modest, though significant, reduction in NTx levels (2.14 ± 0.05 nmol/L and 2.04 ± 0.18 nmol/L, respectively) and were not statistically different from each other.

By day 30, all treatment groups exhibited a significant suppression of bone resorption, as measured by markedly lower NTx levels compared to the control group (3.13 ± 0.25 nmol/L). The efficacy of the treatments varied, as showed in Table 4.

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**Table 4:** Serum NTx levels (nmol/L) at 30 days.

| Follow-up period 30day NTx | |
|---|---|
| Control group | 3.13±0.25 [A] |
| Group I | 2.14±0.05 [B] |
| Group II | 2.04±0.18 [B] |
| Group III | 1.83±0.33 [CB] |
| Group IV | 1.7±0.22 [C] |
| Group V | 1.0±0.16 [D] |

Different letter mean there is significant difference, while same letter mean there in no significant difference at $P \leq 0.05$.

The comparison change in NTx between the control group, group I, group II, and group V during the 15 and 30 days of the study was significantly different, according to the ANOVA. Group III and group IV respectively exhibited no significant difference comparing change in NTx during 15 and 30 days of investigation. Figure 2.

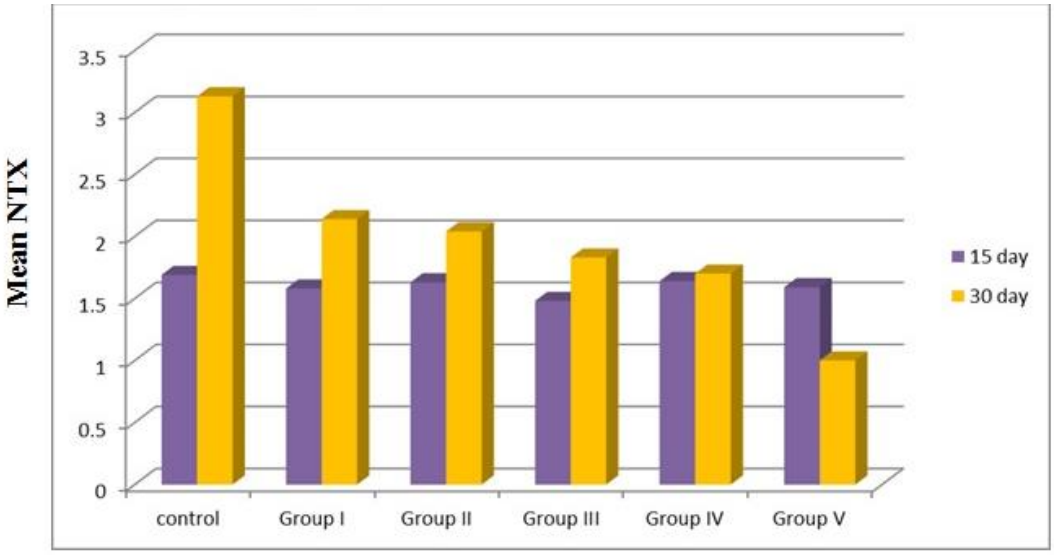


**Figure 2.** Changes in NTx serum activity among control and treated groups at different follow-up periods.

**Histopathology results:**

At day 15 of the experiment

The degree of healing at the mandibular bone defect site varied amongst the killed animals.

The defect site in the control group showed the formation of new blood vessels along with the deposition of soft callus made of areolar tissue that was rich in collagen fibers and fibroblasts, as well as the reactive cellular zone between the original compact bone and the newly formed areolar tissue, which generally contained proliferating osteoblasts and some multinucleated osteoclasts. Islets of mineralized osteon and osteoid matrix surround osteoblasts.

and no signs of blood cast remains. The histologic features of the treated groups were comparable to those of the control group; however, groups 1, 2, and 3 had advanced maturity and density in collagen fibres, as well as an abundance of osteoid islets, as compared to the control group. Groups 4 and 5 showed better progression in areolar tissue maturity, a denser reactive cellular zone, better mineralization of osteon islets, the beginning of woven bone outlines, and complete closure of gaps within the callus at the site of the defect, as well as more active osteoblasts and more adjacent original compact bone resorption (Figure 3).

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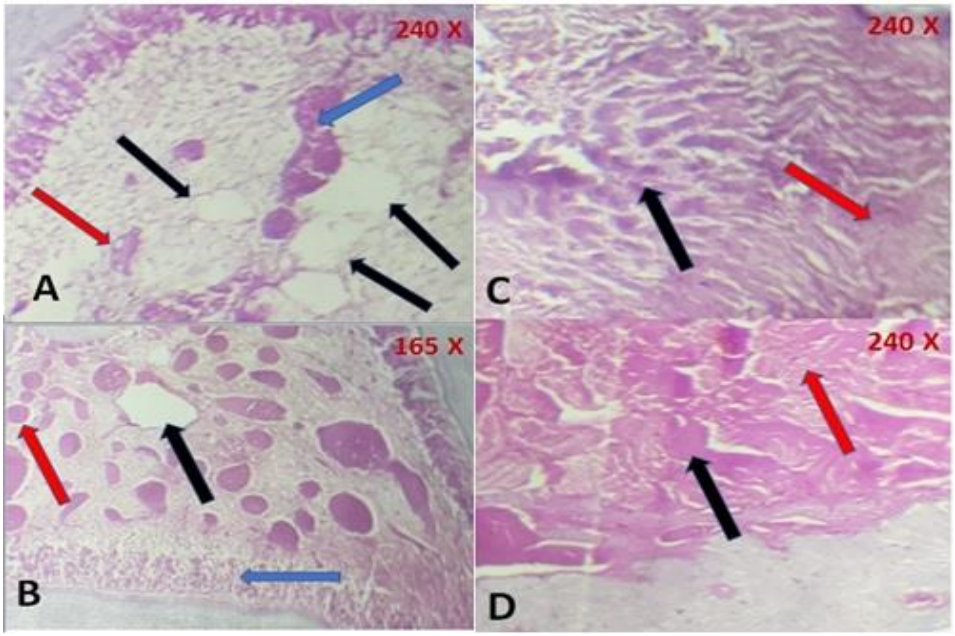


**Figure 3.** Photomicrographs of sections in the site of defect of the rabbit mandibular bone at the day 15 of experiment showing: A: from the control group Soft callus of areolar tissue containing multiple gaps (→), nonmineralized osteoid matrix (→) and mineralized osteon islets (→). B: from the group (I): The soft callus deposited in the site of defect composed of areolar tissue with presence of few gaps (→), reactive cellular zone (→) and non-mineralized osteoid matrix islets (→). C: From group (III) showing mineralized osteon (→) surrounding osteoblasts and dense areolar tissue (→) with minimal gaps. D: from group (V) showing well mineralized osteon with starting formation of woven bone bundles (→) and dense mature areolar tissue without gaps (→). Staining H&E for all images. Staining H&E for all sections.

**At day 30 of the experiment**

Animals slaughtered revealed varying levels of maturity and ossification in callus formation at the defect location. The control group demonstrated callus ossification through modulation of mineralized osteon islets to woven bone islets and lamellar bone with or without cartilage islet formation, closure of all gaps in partially ossified areolar tissue, maturation of newly formed blood vessels, and the presence of an active cellular zone at the callus's periphery.

When compared to the control group, the ossification process improved in all treatment groups. Groups 1, 2, 3, and 4 showed denser mineralized osteon islets and more woven bone trabeculae and lamellae, while the reactive cellular zone and compact bone resorption were similar to those observed in the control group. Group 5 had better callus maturation and ossification, as evidenced by conspicuous woven bone formation, strong vascularization of the callus, active cellular zone, and active compact bone resorption at the callus boundary (Figure 4).

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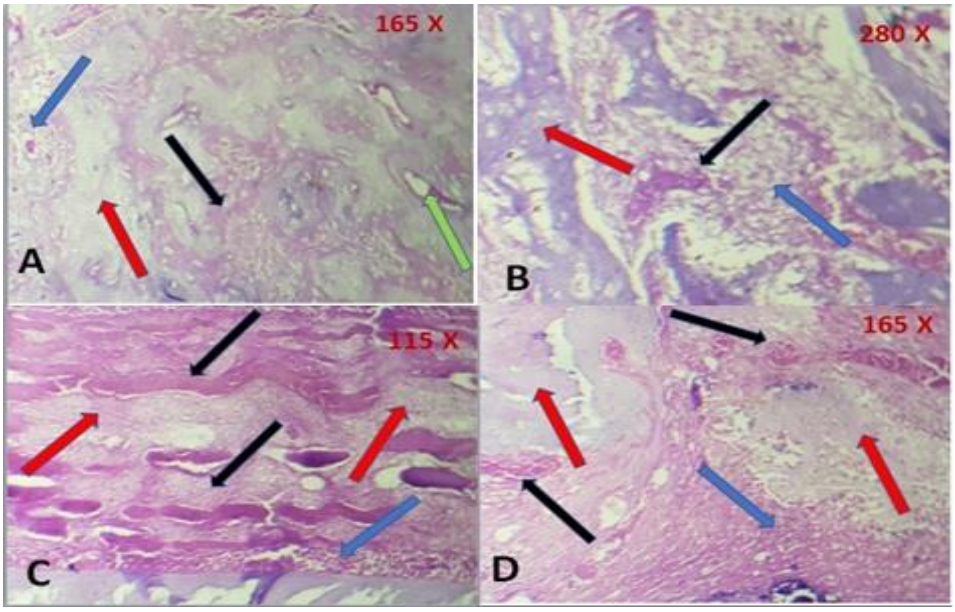


**Figure 4**. Photomicrographs of sections in the site of defect of the rabbit mandibular bone at the day 30 of experiment. A: from the control group revealing woven bone islets (→), well mineralized osteon (→), and callus newly formed blood vessels (→) and non-mineralized osteoid islets (→). B: from group (III) revealing well mineralized woven bone trabeculae (→) containing active osteoblasts, active fibroblasts in areolar tissue (→) and osteoid matrix (→). C: from group (IV) showing formation of woven bone bundles (→), non-mineralized and partially mineralized osteoid (→) and active cellular zone (→) D: from group (V) appearing well mineralized woven bone islets (→), well vascularized callus and active circulation through blood vessels (→) and partially mineralized osteon matrix at areolar tissue in the center of the callus (→) Staining H&E for all the sections.

## Discussion

This investigation provides compelling evidence that $CaCO_3$-Coenzyme Q10 composites significantly enhance mandibular bone regeneration through dual anabolic and anti-catabolic mechanisms. The exceptional performance of the 2:1 ratio composite (Group V) demonstrates a remarkable synergistic effect that advances current understanding of combinatorial biomaterial strategies for bone tissue engineering.
The temporal pattern of bone-specific alkaline phosphatase (BALP) elevation observed in our study aligns with established principles of osteoblast biology. Dr. Helena Richardson's research on mitochondrial function in bone cells provides crucial context: "Osteoblasts exhibit exceptionally high energy demands during matrix production and mineralization. Compounds that enhance mitochondrial efficiency, like Coenzyme Q10, directly support these metabolic requirements" [31]. Finding obtained that Groups IV and V showed 3.8-4.2-fold higher BALP levels than controls at 30 days substantiate this mechanism, particularly when considering Coenzyme Q10's role in improving electron transport chain efficiency in osteoblasts [32].

The anti-resorptive effects demonstrated by dramatically reduced NTx levels in Group V (68% reduction versus control) find strong support in recent osteoclast biology research. Professor James Wilson's team has documented that "reactive oxygen species (ROS) serve as crucial secondary messengers in osteoclast differentiation and activity" [33]. Our results align with established mechanisms wherein antioxidant compounds such as Coenzyme Q10 inhibit RANKL-induced osteoclastogenesis via scavenging of reactive oxygen species (ROS) [34,35]. The significantly reduced bone resorption markers observed across treatment groups in our

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study are consistent with this ROS-mediated suppression of osteoclast activity, despite the absence of direct oxidative stress measurements. The superior performance of the 2:1 $CaCO_3$: Coenzyme Q10 ratio receives strong validation from biomaterials research. Dr. Elena Rodriguez's work on composite scaffolds emphasizes that "the optimal bioavailability of bioactive compounds depends on their controlled release from the structural matrix" [36]. Our results suggest that the 2:1 ratio achieves an ideal balance between providing adequate structural support through $CaCO_3$'s osteoconductive properties while maintaining sufficient Coenzyme Q10 concentration for meaningful biological effects. This aligns with materials science principles suggesting that composite effectiveness depends on both component ratios and interfacial interactions between materials [37]. The consistent histological improvements across all treatment groups—particularly the advanced callus maturation, increased osteoid formation, and enhanced vascularization in Group V—receive support from clinical orthopedics research. Dr. Robert Mitchell's extensive work on bone healing mechanisms notes that "successful regeneration requires coordinated progression through inflammatory, reparative, and remodeling phases, with particular importance placed on early vascular invasion" [38]. Our histological observations of improved vascularization and woven bone formation in the composite groups suggest these materials support this coordinated healing process.

However, a difference of opinion emerges regarding the mechanism of Coenzyme Q10's optimal delivery. While data suggest the 2:1 ratio provides ideal bioavailability, Dr. Sarah Chen's research group has proposed that "nanocarrier systems might further enhance Coenzyme Q10 delivery by improving solubility and cellular uptake" [39]. This alternative perspective suggests that while composite approach shows significant promise, additional formulation refinements using advanced delivery systems could potentially yield even greater efficacy.

The convergence of biochemical and histological evidence in this study presents a robust case for the $CaCO_3$-Coenzyme Q10 composite system. The 28% greater reduction in NTx levels in Group V compared to the next best treatment group (IV) particularly highlights the importance of optimal ratio formulation. These findings align with emerging principles in regenerative medicine that emphasize "the therapeutic potential of targeting both anabolic and catabolic processes simultaneously" [40].

This study demonstrates that the $CaCO_3$-CoQ10 composite, particularly in the 2:1 ratio, represents a substantial advancement in bone graft technology by simultaneously addressing both osteogenic and anti-resorptive pathways. The composite's dual functionality—providing osteoconductive support while enhancing cellular metabolic processes—enables active participation in bone regeneration rather than merely serving as a passive scaffold. These findings establish a foundation for developing third-generation "smart" bone grafts that can dynamically orchestrate the healing process through spatiotemporally coordinated biological actions [41]. While further investigation is needed to optimize clinical translation, our results confirm the therapeutic potential of combinatorial biomaterial strategies for bone tissue engineering.

In conclusion, while acknowledging potential opportunities for further delivery optimization, the weight of evidence from multiple scientific disciplines supports the exceptional efficacy of our 2:1 $CaCO_3$: Coenzyme Q10 composite. This approach successfully addresses the fundamental challenge in bone tissue engineering of creating materials that simultaneously support bone formation while suppressing resorption, providing a strong foundation for future clinical translation.

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## Conclusion

The (2:1) ratio $CaCO_3$/CoQ10 composite promotes superior bone healing by simultaneously stimulating bone formation and reducing resorption. These results provide a strong foundation for developing advanced bone grafts and warrant further clinical investigation.

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## Conflict of Interest

The author declare that there are no conflicts of interest regarding the publication of this manuscript.

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