

Teriparatide as an Adjunct to Bovine Bone Grafting for the Repair of Critical-Sized Bone Defects in Rats



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INTRODUCTION

Bone grafting is the treatment of choice for osseous defect repair in maxillofacial areas due to pathologies including periodontitis and tooth loss. There is a need to further develop and optimize bone graft materials to aid in bone regeneration. Previous studies have shown that bovine bone (BB) expresses osteoconductive properties, as well as that intermittently administered parathyroid hormone (PTH) has anabolic effects on bone. We hypothesized that impregnation of BB with PTH will result in enhanced bone formation in the treatment of critical-size bone defects (CSBDs), in terms of bone volume and bone quality, as compared to BB alone.

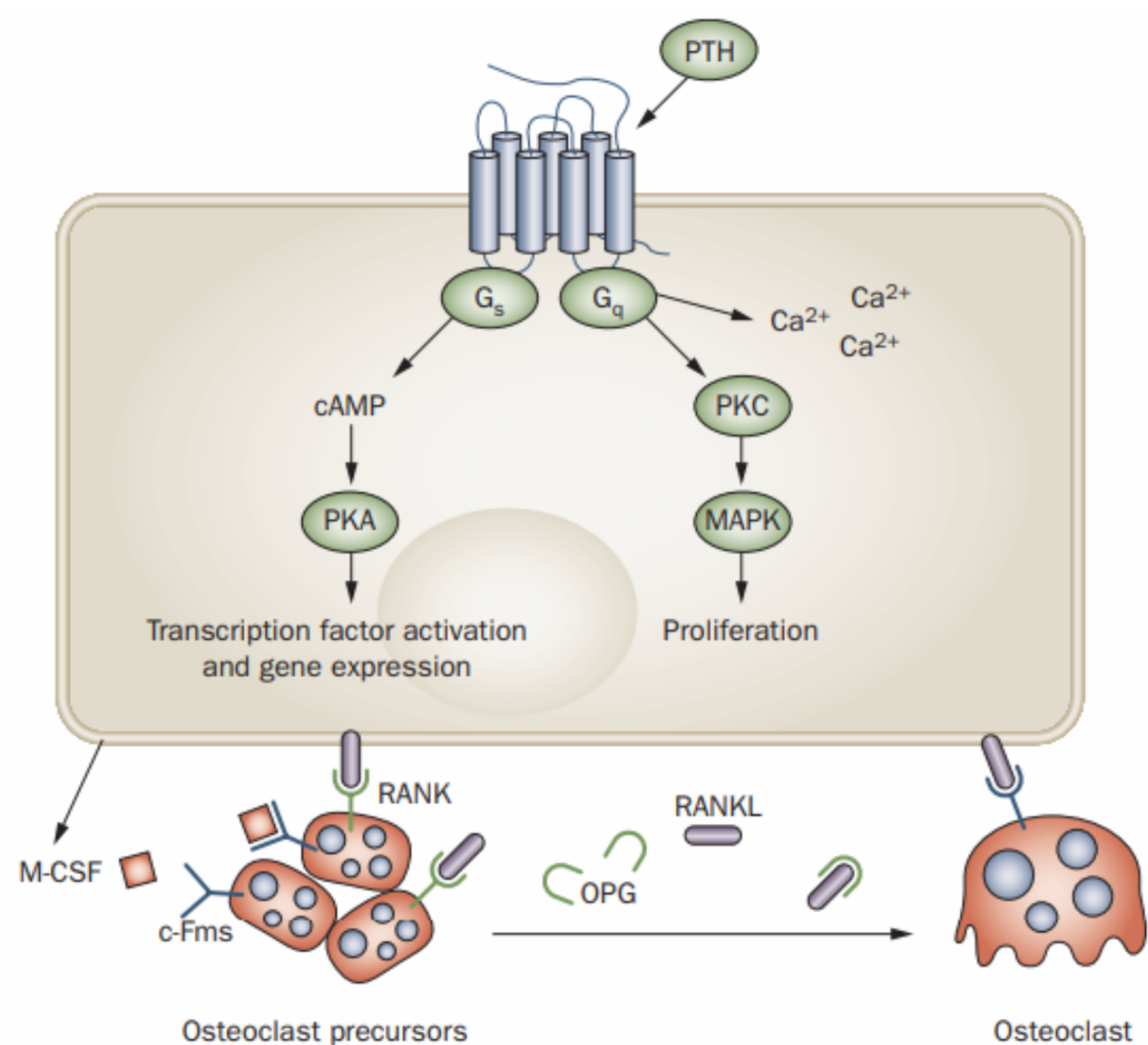
AIM

To determine if the release of PTH1-34 (T) from the impregnated BB particles and/or subcutaneous (SC) administration of T positively modulates bone homeostasis, remodeling, micro architecture, and new bone formation.

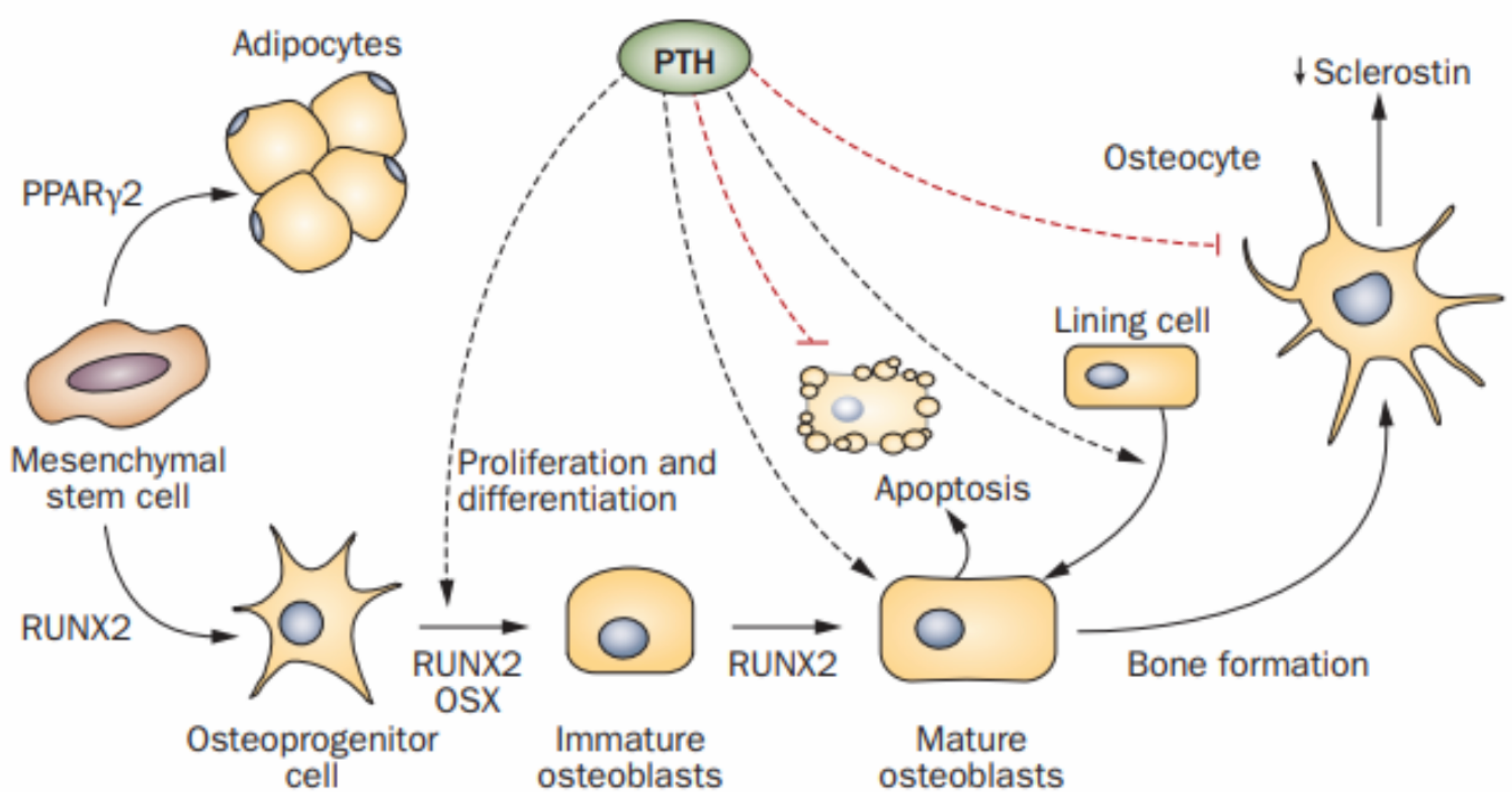
WHAT IS TERIPARATIDE?

PTH is a peptide of 84 amino acids secreted by the parathyroid glands
The recombinant form of PTH1-34, named Teriparatide (T), is the most commonly used FDA approved, anabolic bone drug to treat severe osteoporosis. Chan HL and McCauley LK, 2013

CATABOLIC EFFECTS



ANABOLIC EFFECTS



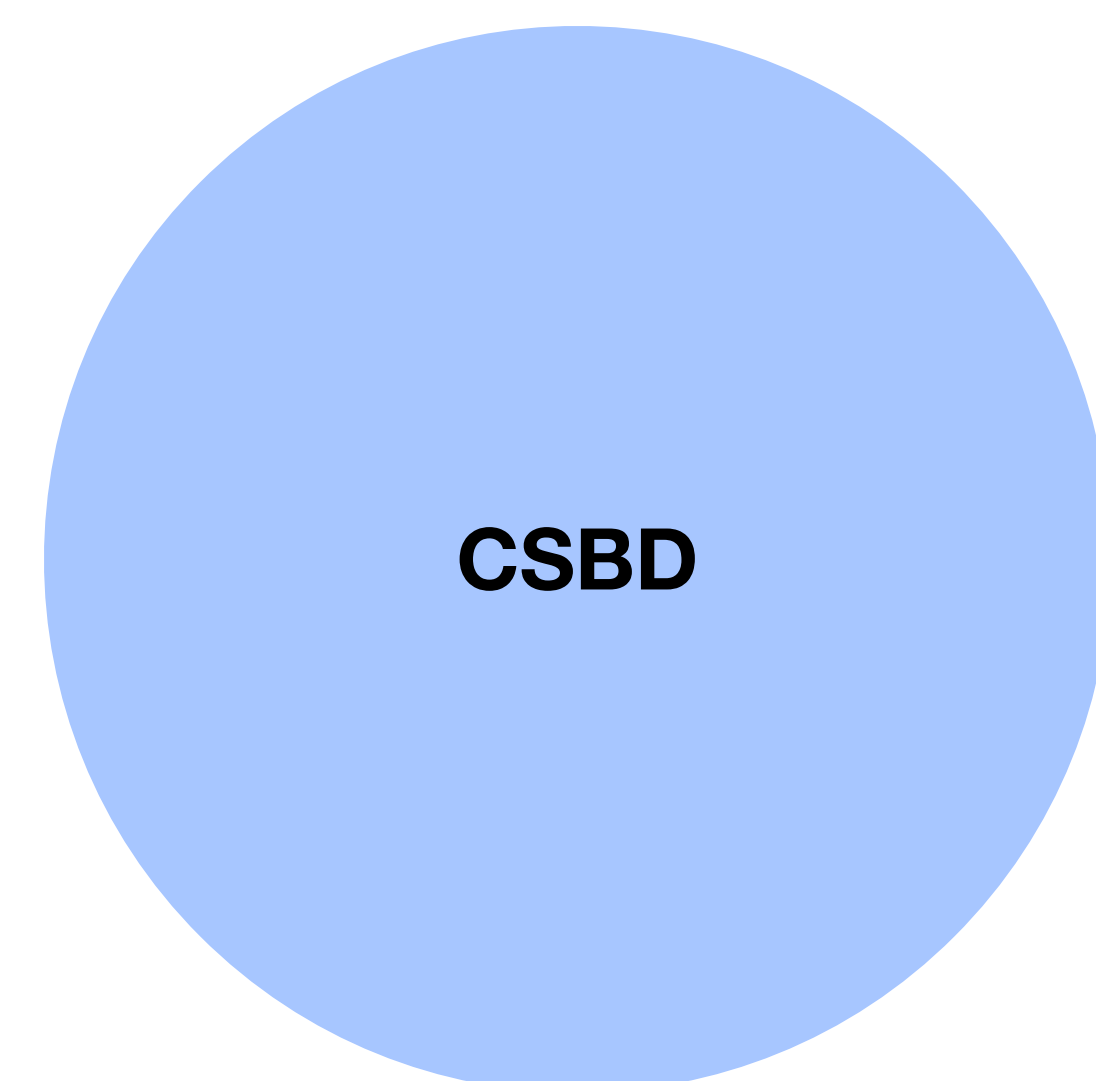
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METHODS

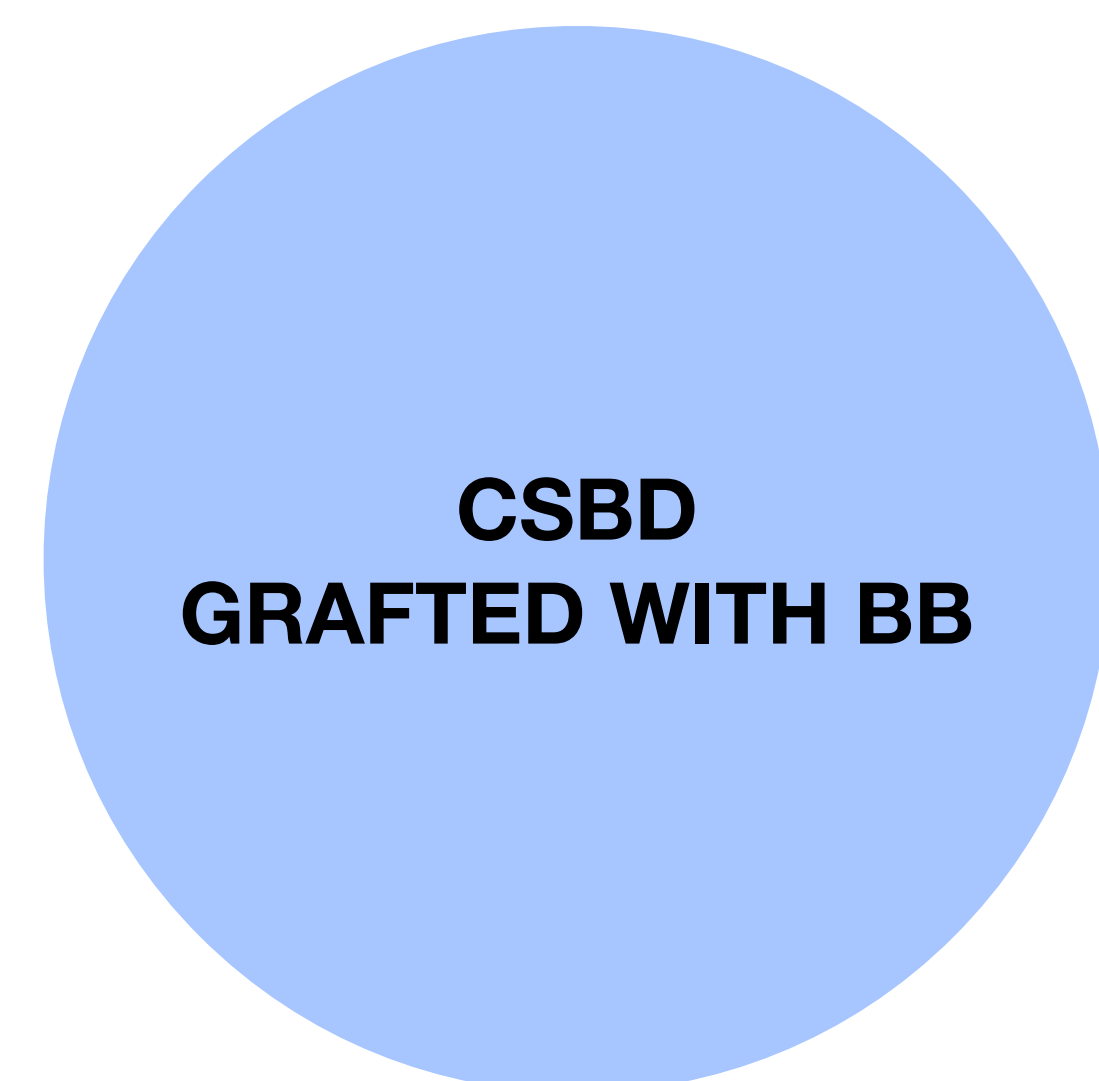
EXPERIMENTAL DESIGN

28 Adult male Wistar rats were assigned to the following groups (N=7/ group)

CONTROL



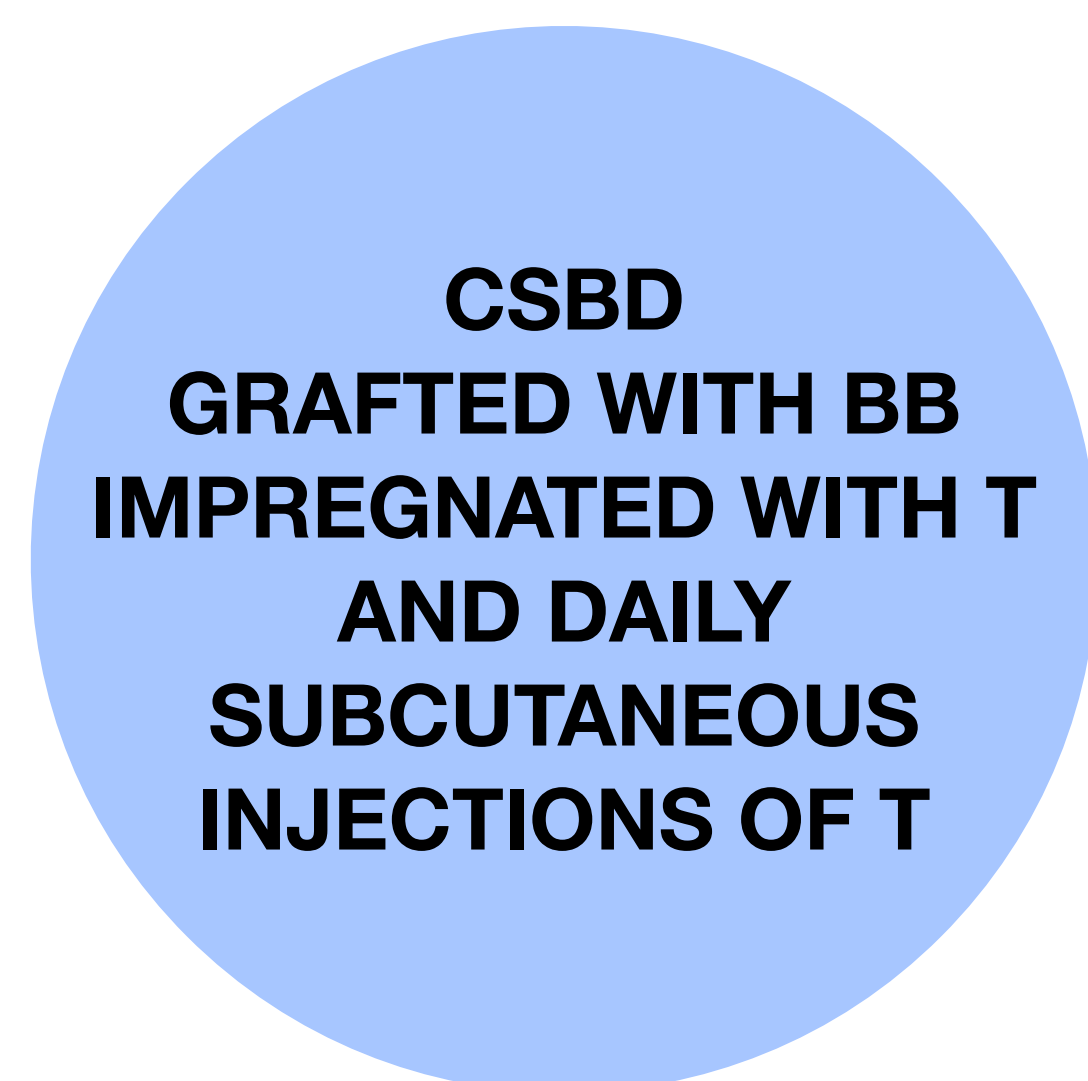
BB



BB & T

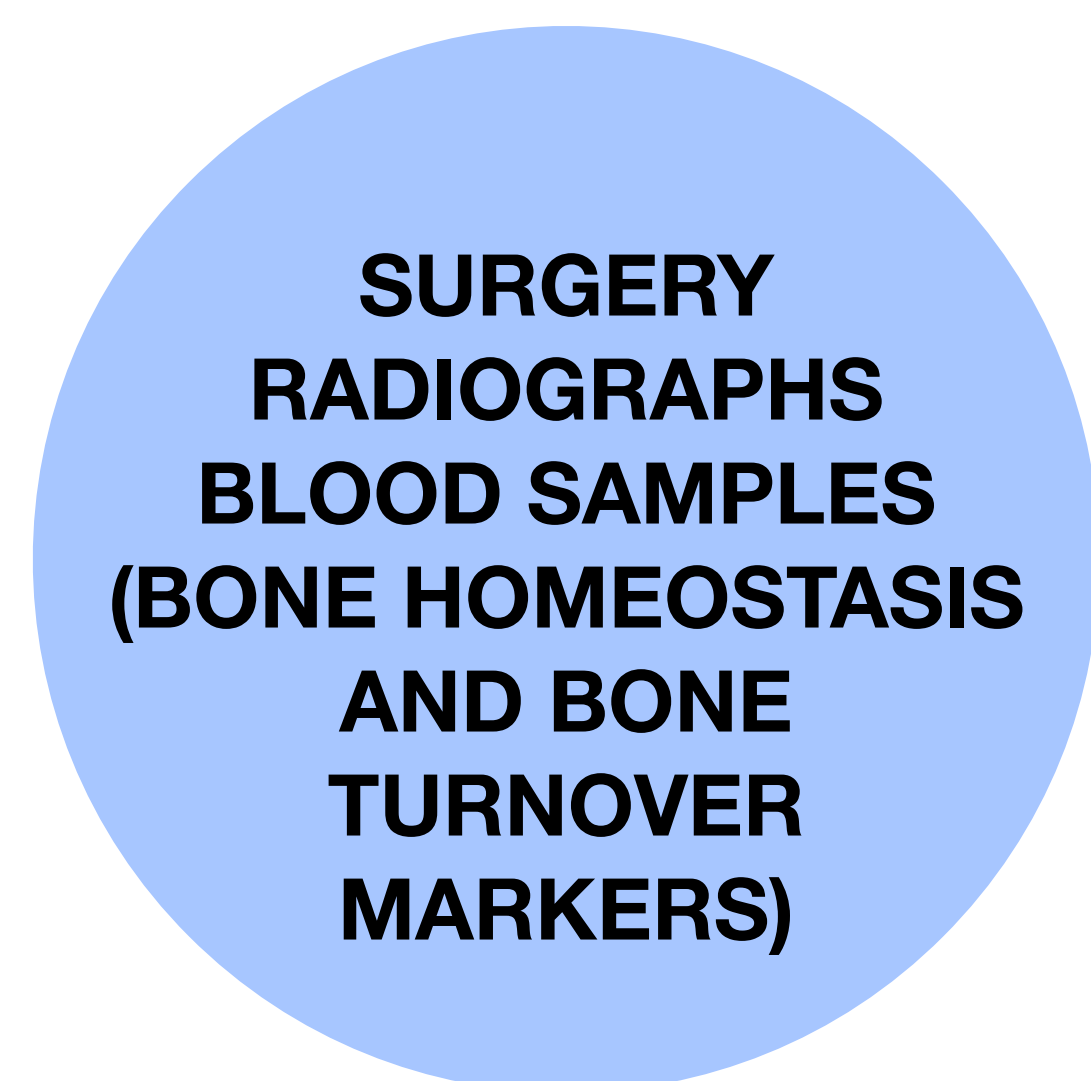


BB & T + T SC

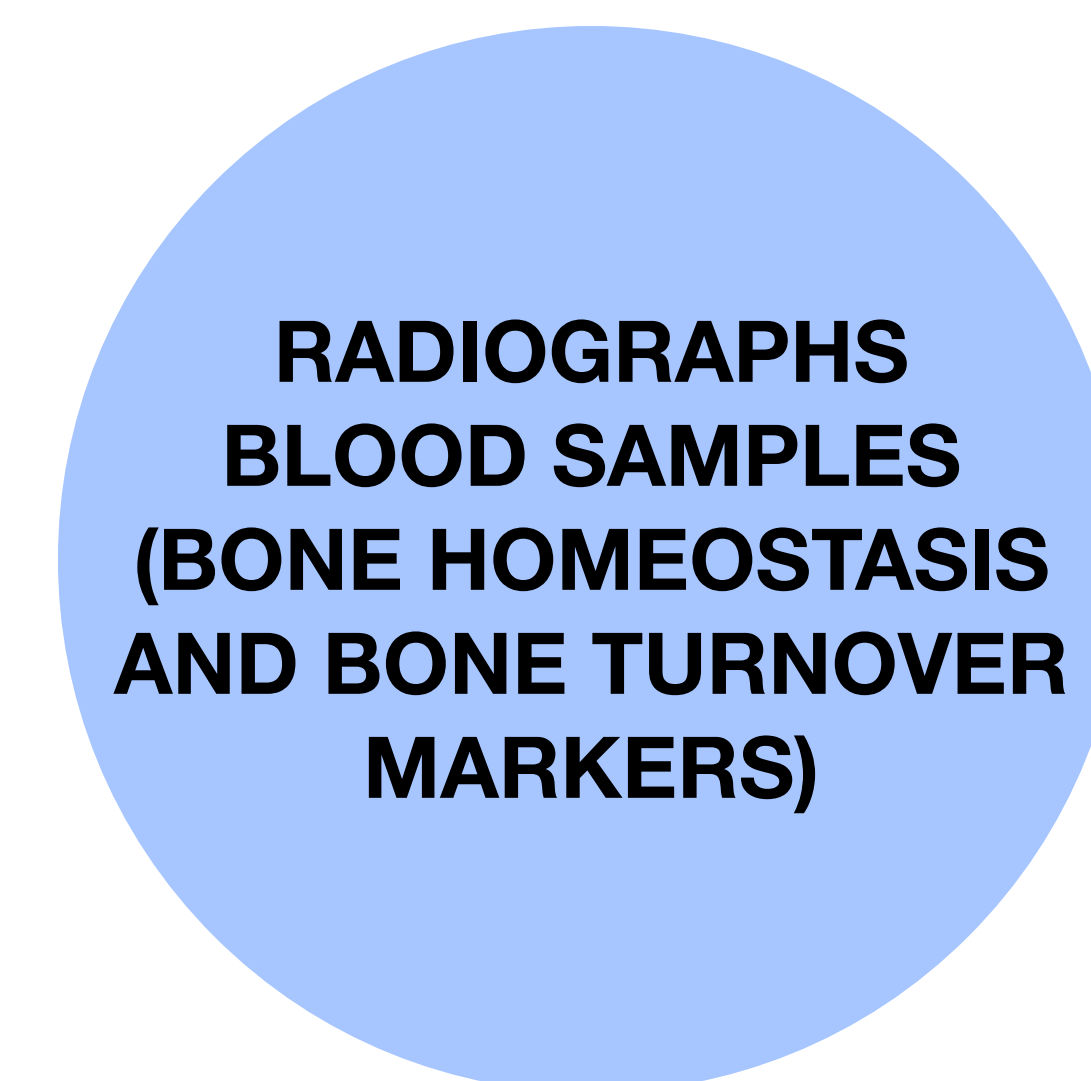


Timeline

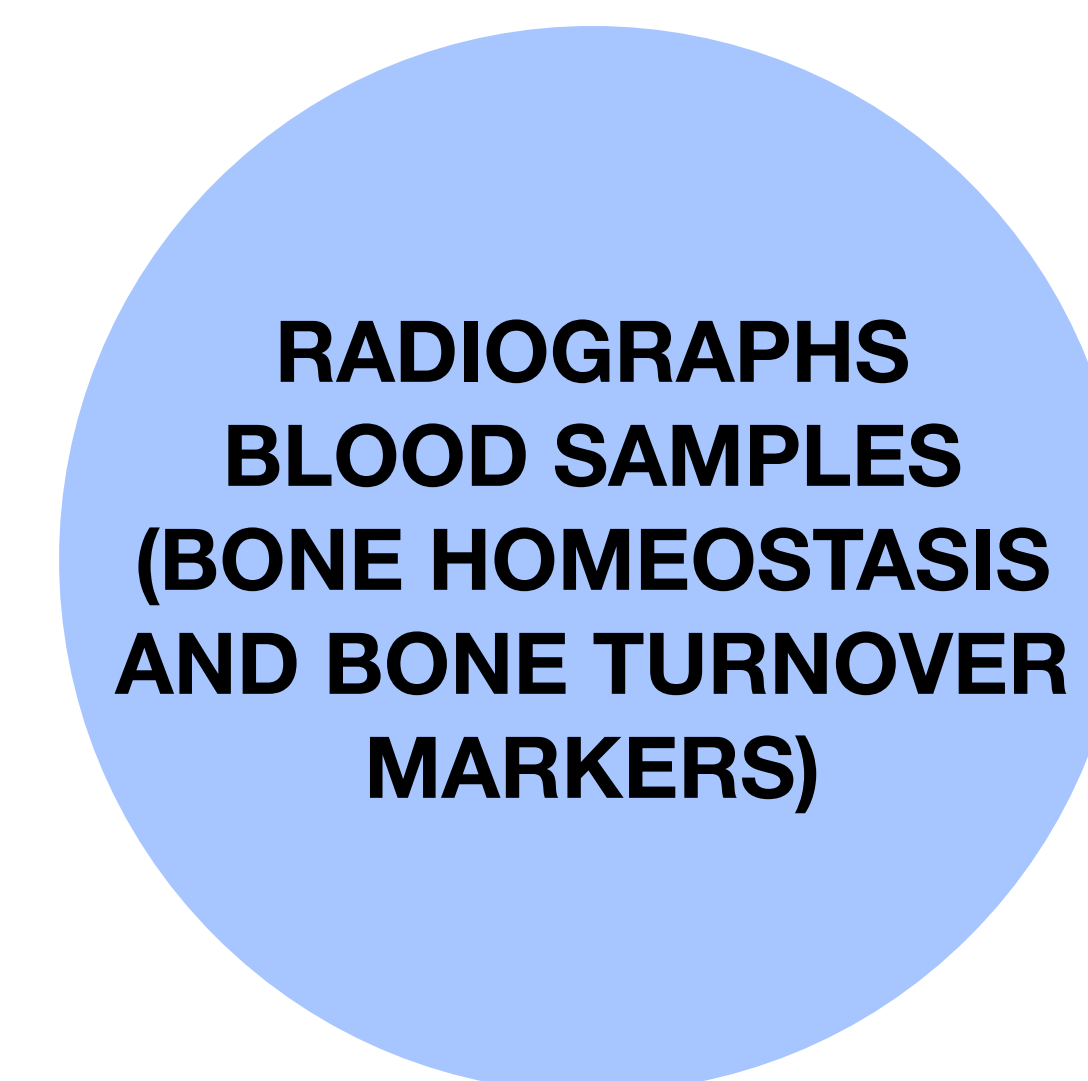
T = 0



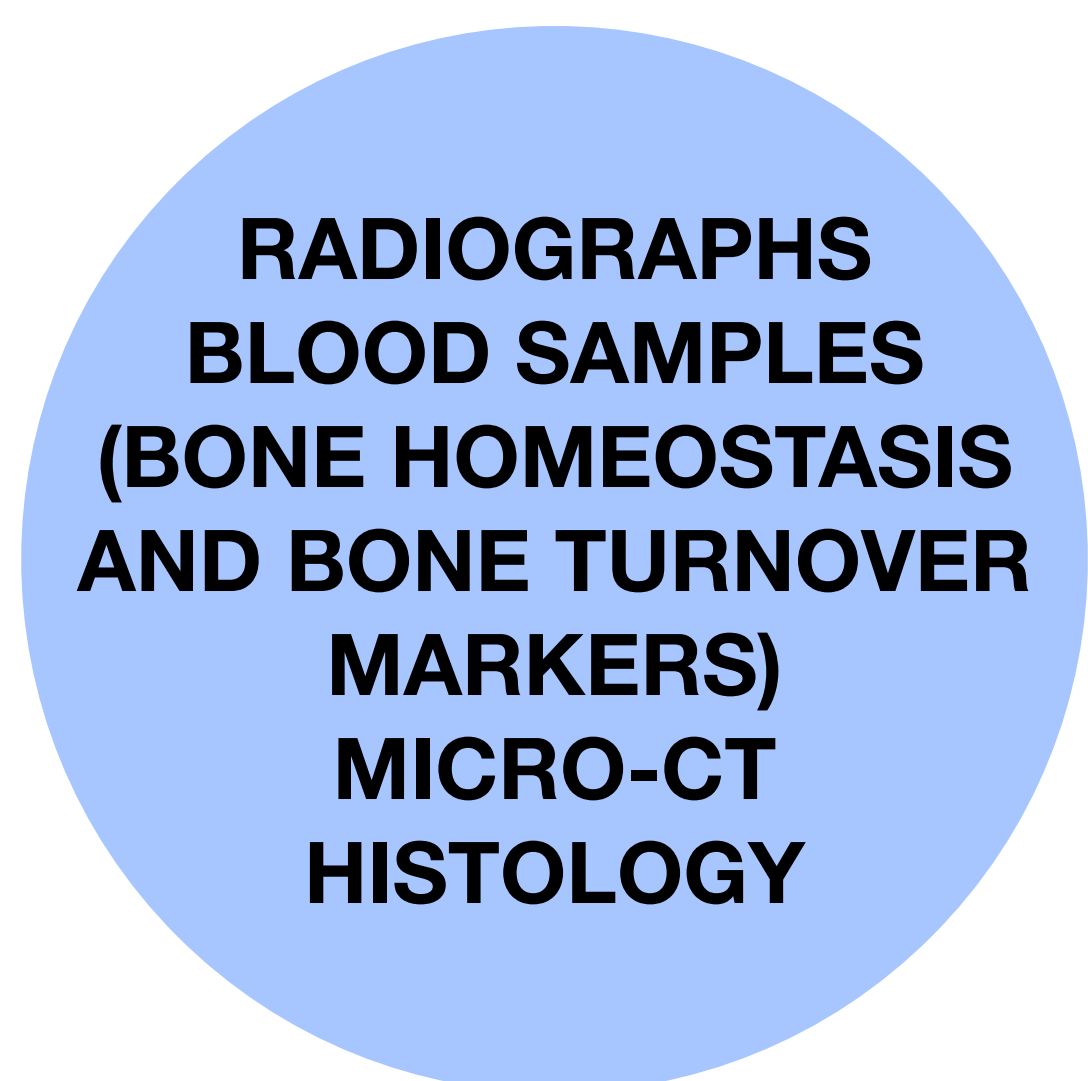
T = 15D



T = 30D



T = 45D



SURGICAL PROCEDURES

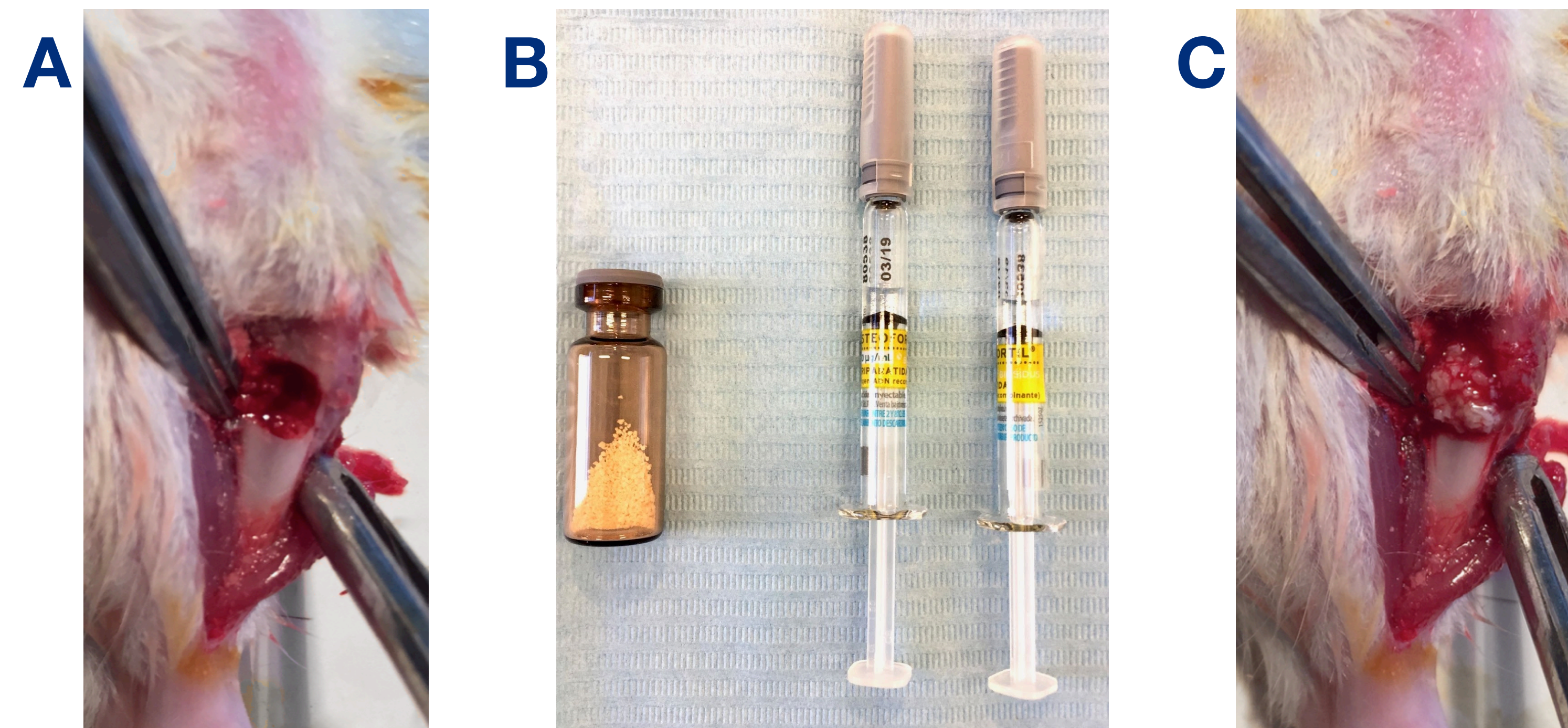


Figure A
Critical-sized bone defect in the mesial aspect of the tibia

Figure B
Bovine bone graft (Synergy, Odontit, Argentina) and T solution (Osteofortil, Biosidus, Argentina)

Figure C
Critical-sized bone defect grafted

ANALYSES

Biochemical analyses

Bone homeostasis: PTHi by ELISA (MyBioSource, San Diego, USA)

Bone formation markers: N-terminal propeptide of type I collagen (P1NP) and osteocalcin (OCN) by ELISA (MyBioSource, San Diego, USA)

Bone resorption marker: C-terminal telopeptide of type I collagen (CTX) by ELISA (MyBioSource, San Diego, USA)

Micro-CT analysis

Bruker SkyScan 1272 (Bruker Corporation, Kontich, Belgium) at 70 kV, 142 μ A, and a resolution of 25 μ m, 180° rotation around the vertical axis, and a rotation step of 0.7° using a 0.5-mm-thick aluminum filter and a 0.038-mm-thick copper filter. Images were reconstructed using NRecon v.1.7.3.1 software (Bruker-microCT)

Histomorphometry

% BV/TV evaluated using Image-Pro

Statistical analyses

Means $\pm$ standard deviations (SD) are reported. * $p < 0.05$ assessed by one-way ANOVA after Bonferroni corrections.

Statistical analysis was performed using SPSS (IBM SPSS Statistics, Version 20, IBM Corp) Chicago, IL)

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RESULTS

CLINICAL EVALUATION

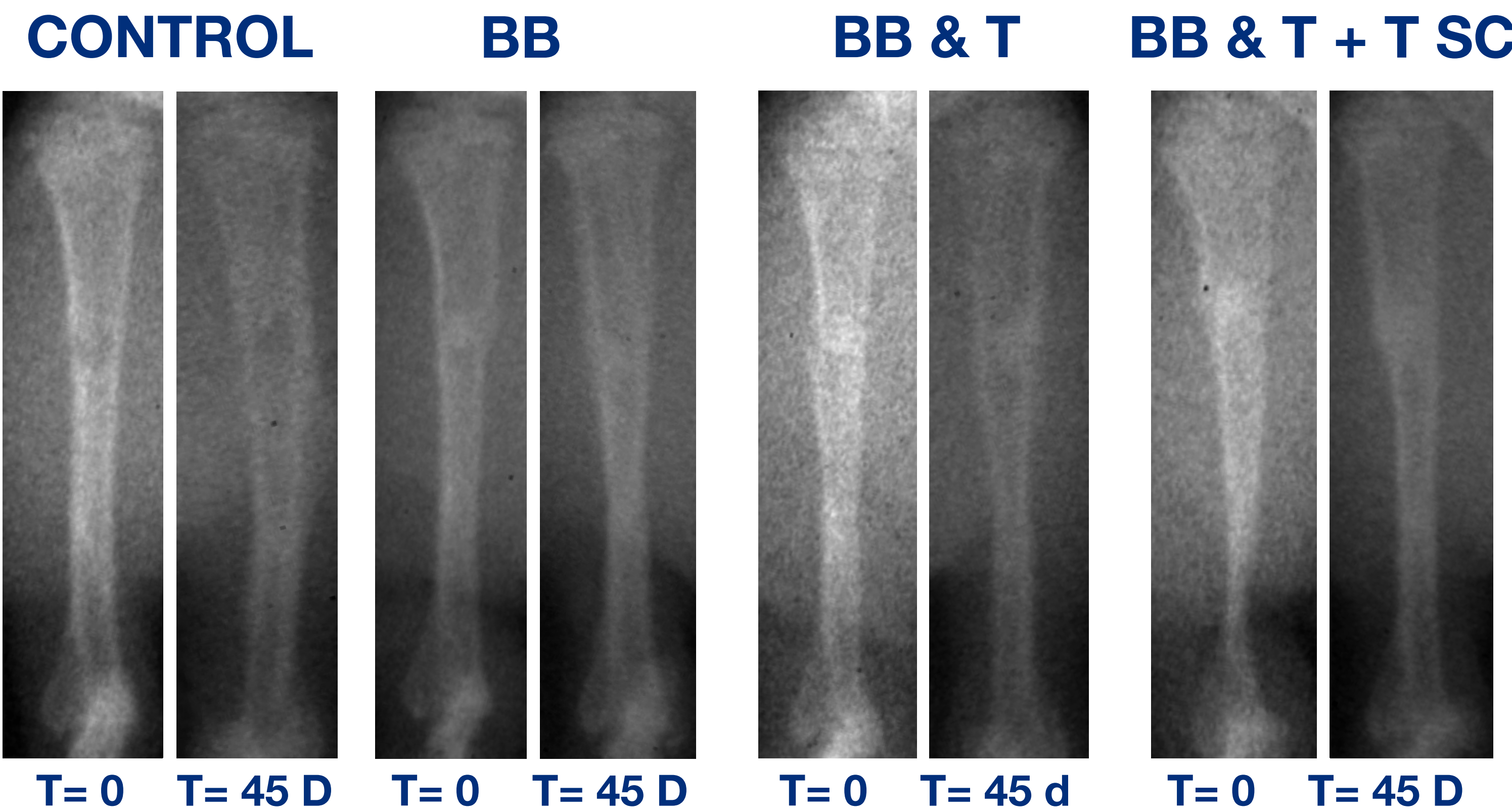
Hematoma					Abscess				
Time	T= 0	T= 15 d	T= 30 d	T= 45 d	Time	T= 0	T= 15 d	T= 30 d	T= 45 d
Control	-	-	-	-	Control	-	-	-	-
BB	-	-	-	-	BB	-	-	-	-
BB & T	-	-	-	-	BB & T	-	-	-	-
BB & T + T SC	-	-	-	-	BB & T + T SC	-	-	-	-

Lymphadenopathy					Encapsulation				
Time	T= 0	T= 15 d	T= 30 d	T= 45 d	Time	T= 0	T= 15 d	T= 30 d	T= 45 d
Control	-	-	-	-	Control	-	-	-	-
BB	-	-	-	-	BB	-	-	-	-
BB & T	-	-	-	-	BB & T	-	-	-	-
BB & T + T SC	-	-	-	-	BB & T + T SC	-	-	-	-

Edema					Suppuration				
Time	T= 0	T= 15 d	T= 30 d	T= 45 d	Time	T= 0	T= 15 d	T= 30 d	T= 45 d
Control	+	+	-	-	Control	-	-	-	-
BB	++	+	-	-	BB	-	-	-	-
BB & T	+++	+	-	-	BB & T	-	-	-	-
BB & T + T SC	+++	+	-	-	BB & T + T SC	-	-	-	-

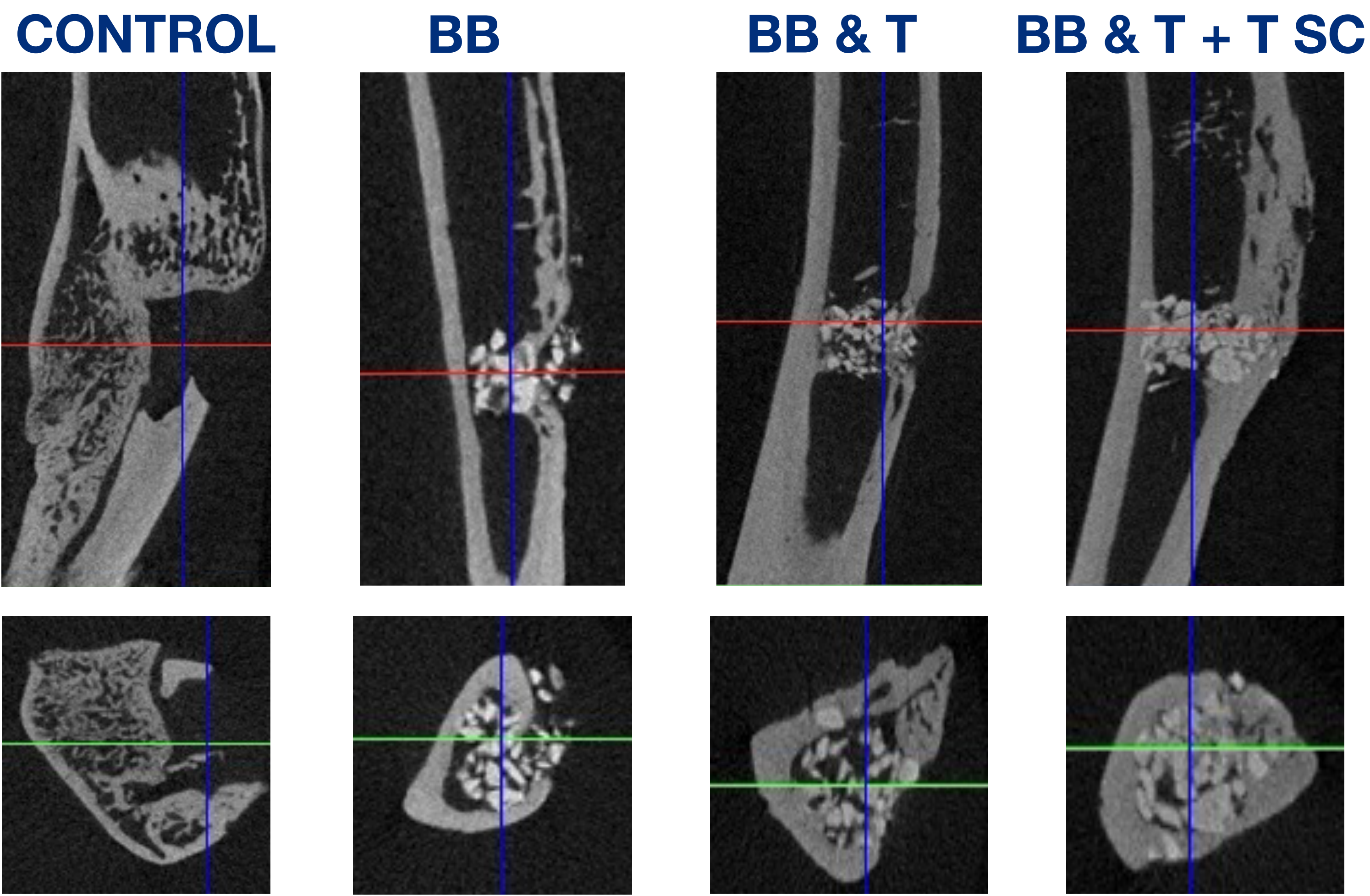
No adverse events were observed at the studied times

RADIOGRAPHIC EVALUATION



An increase in radiopacity was observed at the CSBD in the treated groups at T= 45 d

MICRO-CT EVALUATION

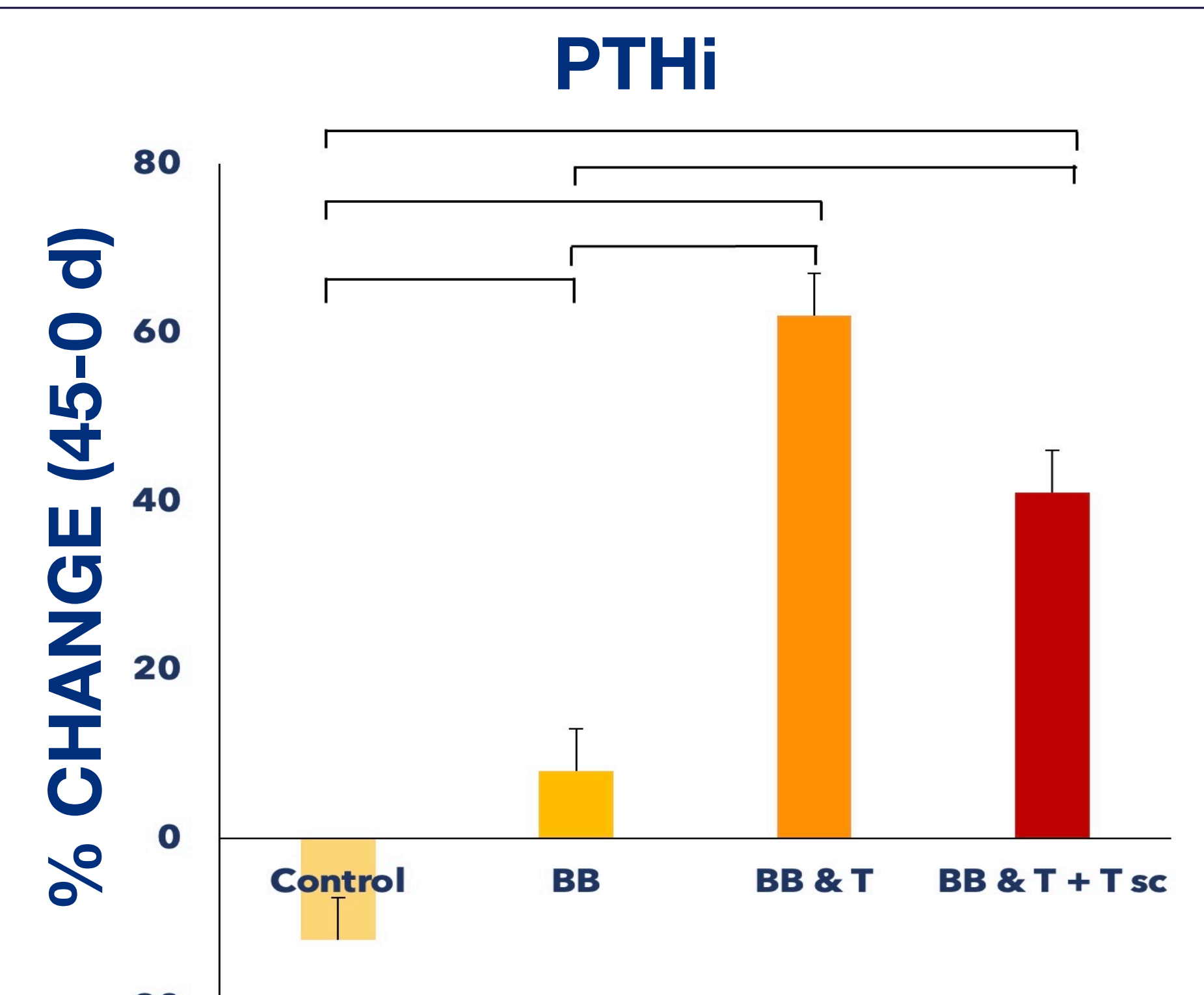


Partial healing of the cortical bone was observed in the BB & T group, whereas complete healing was observed in the BB & T + T SC group at T= 45 d

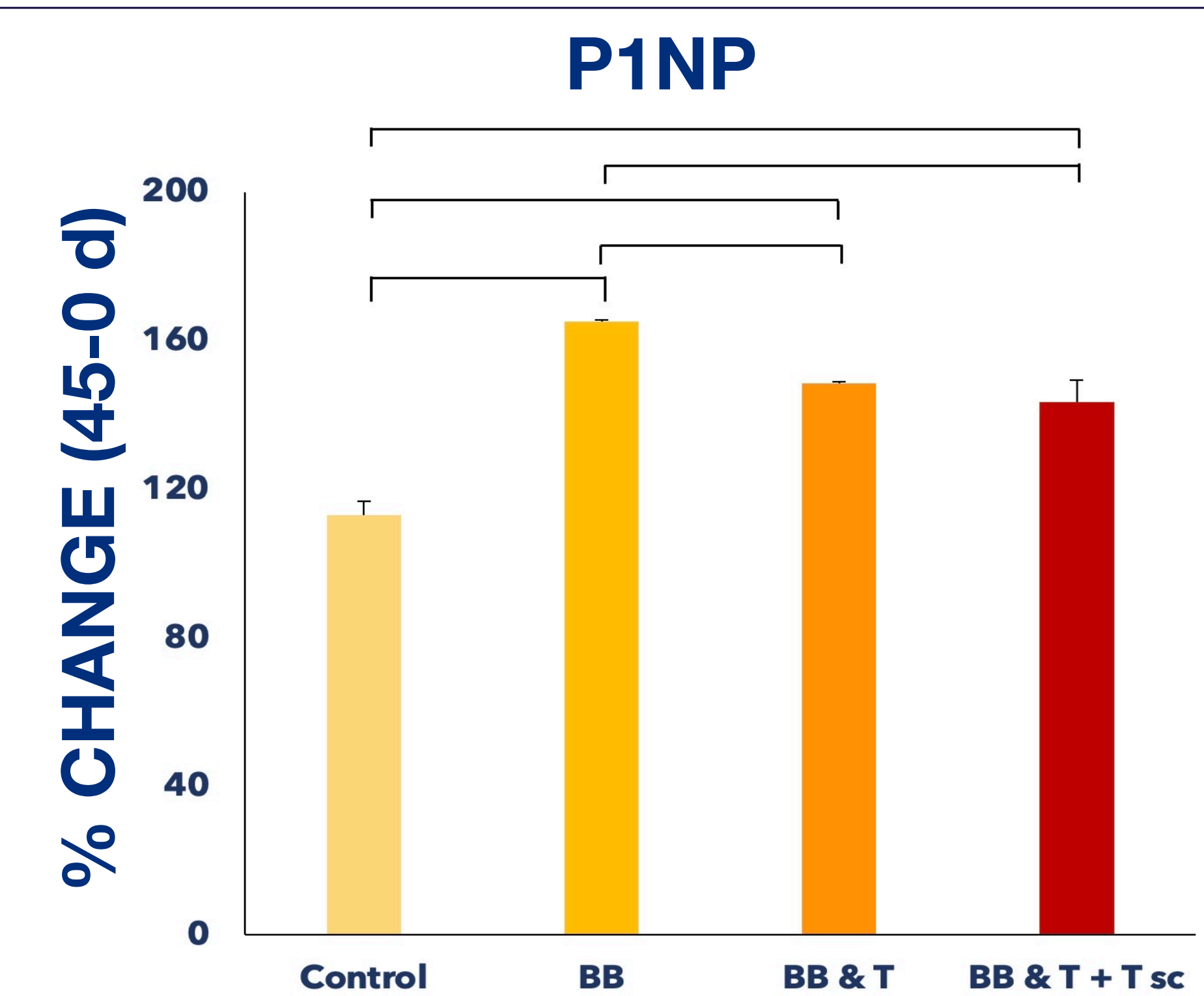
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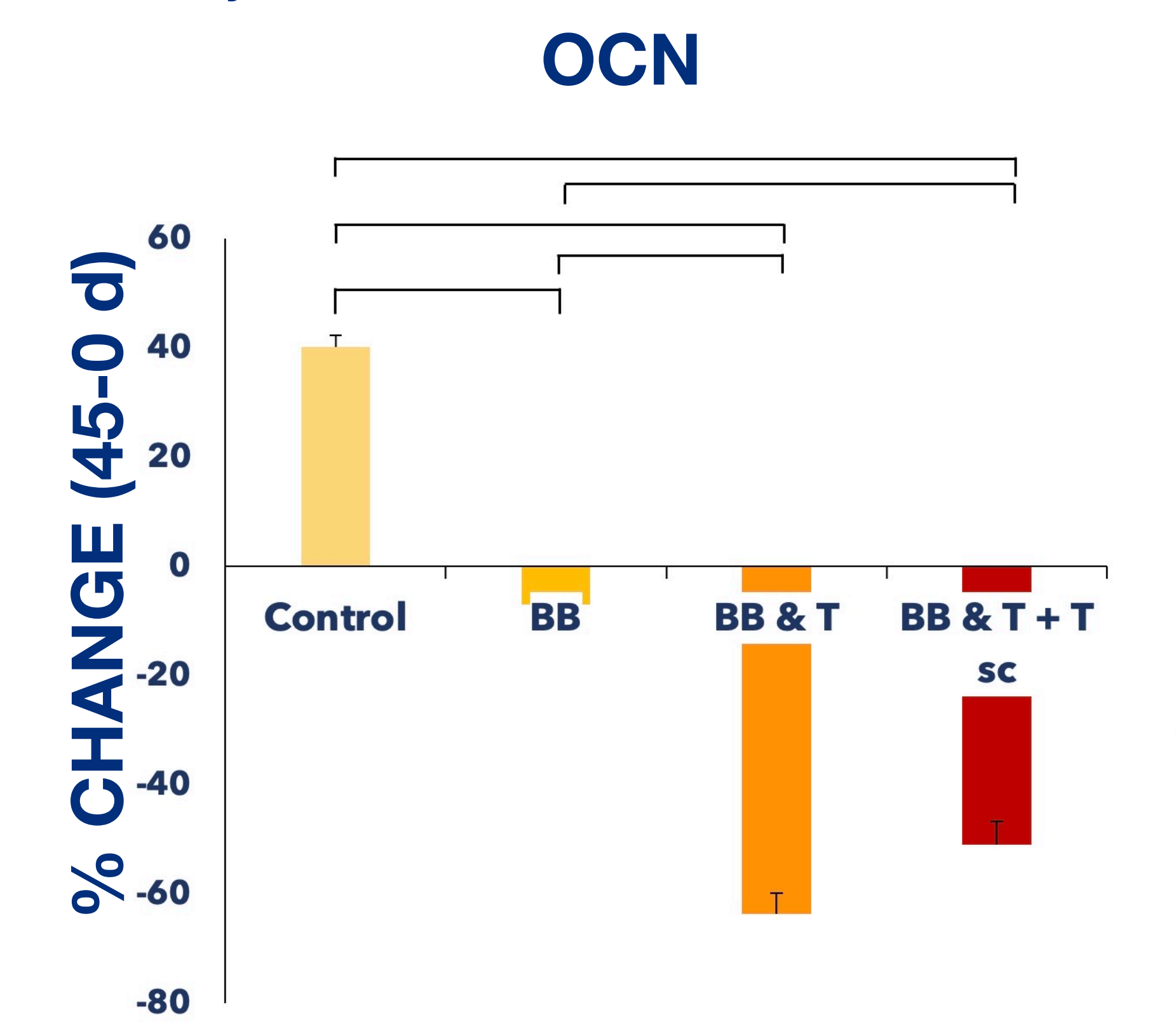
BIOCHEMICAL ANALYSIS



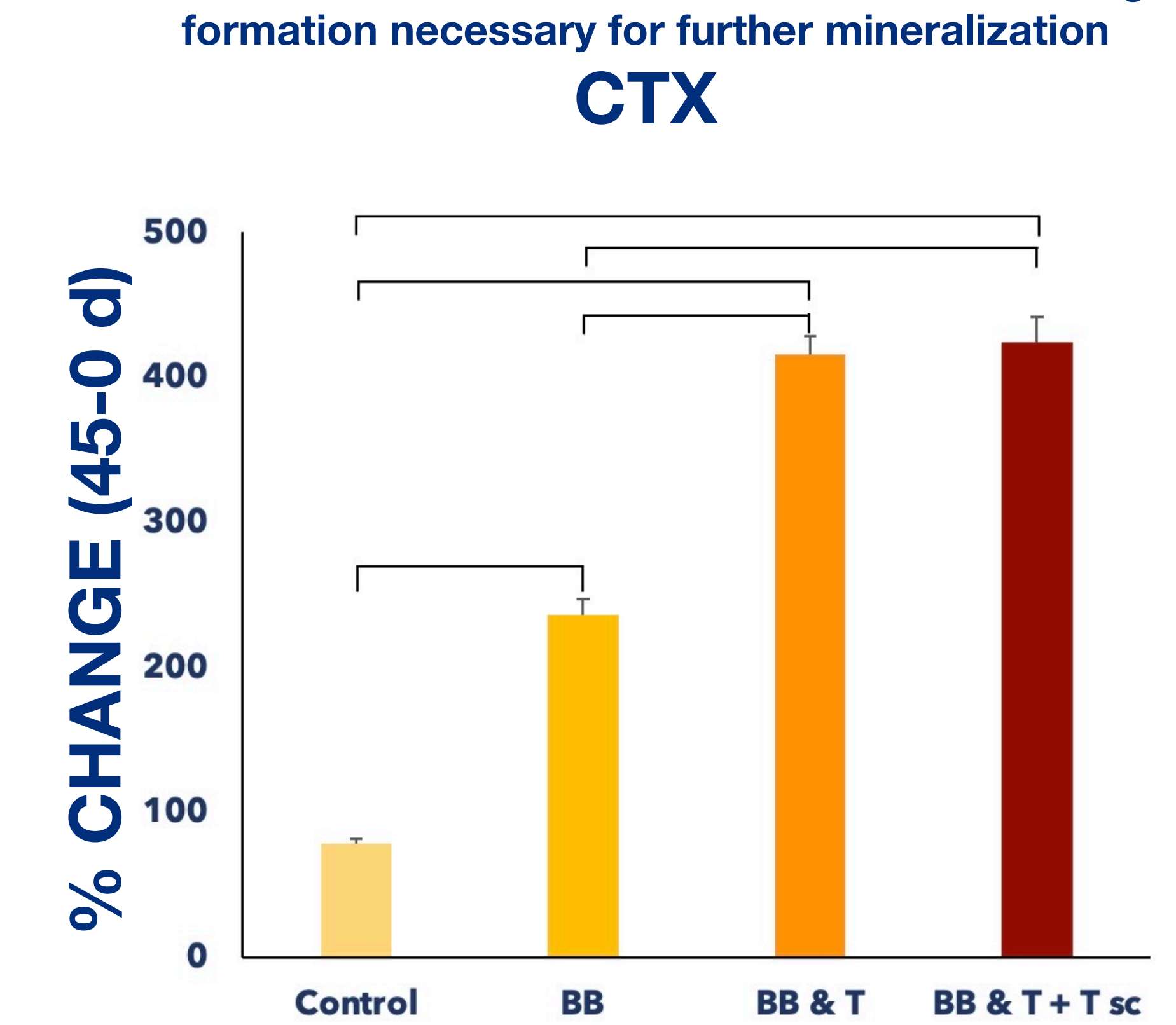
Local release and systemic administration of T increased PTHi levels in serum



The bone formation marker P1NP increased over time indicating active osteoid formation necessary for further mineralization



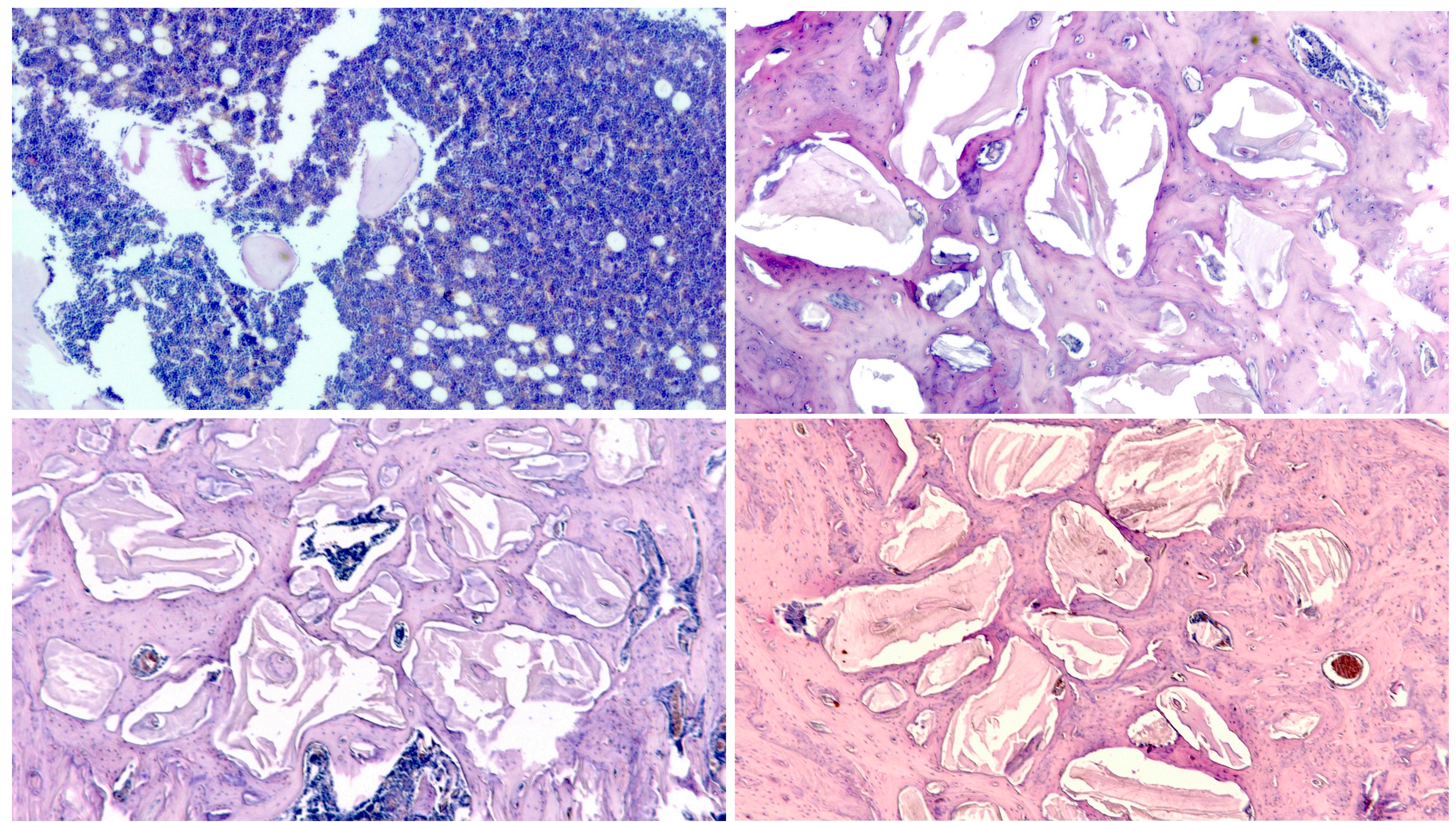
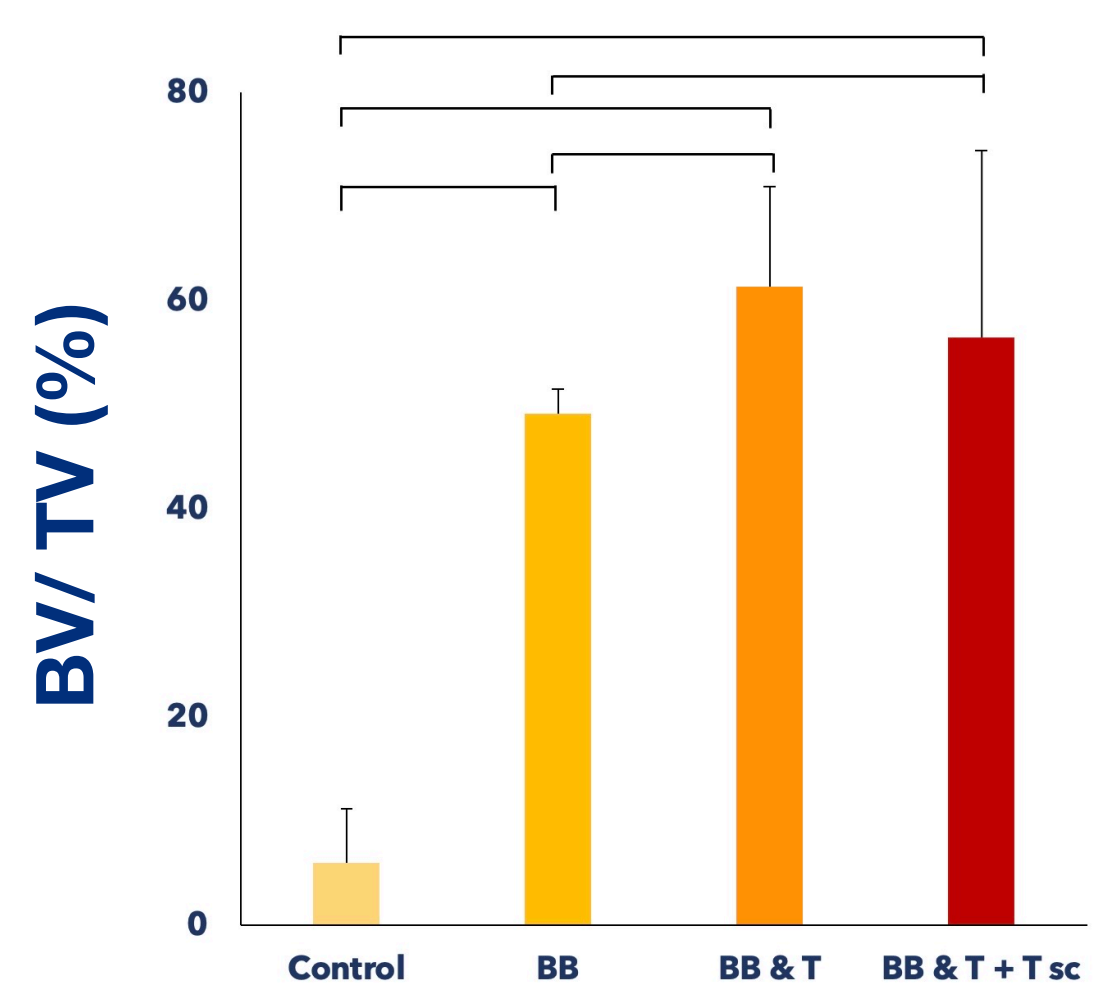
Local release of T as well as systemic administration and local release of T, decreased the osteoblast maturation marker OCN. This decreased is necessary to initiate the bone mineralization process



CTX increased over time indicating the resorption of BB necessary for further replacement with native bone

*p<0.05 assessed by one-way ANOVA after Bonferroni corrections

HISTOLOGY



There was an increase in bone volume in the groups treated with T at T= 45 d

*p<0.05 assessed by one-way ANOVA after Bonferroni corrections

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SUMMARY OF THE RESULTS

Under our experimental conditions

- No adverse effects were observed in the groups that received BB impregnated with T
- The increase in the bone resorption marker CTX, observed in the groups that received T, is likely associated with the degradation of the BB particles for further replacement by native bone
- The decrease in osteoblastic proliferation observed in the groups that received BB impregnated with T indicates an increase in the mineralization of the osteoid. These effects were more pronounced in the group that additionally received systemic T
- Histological and micro-CT evaluation showed that the impregnation of BB with T, lead to partial healing of the cortical at the site of the CSBD, whereas, the addition of systemic T (daily injections of T sc) allowed the complete healing of the cortical bone

CONCLUSION

Our findings indicate that the addition of T to BB enhanced the bone repair process. The systemic and local effects of the combination of BB with T may facilitate a novel therapeutic alternative for the repair of bony defects in the oral cavity.

ACKNOWLEDGMENTS

Supported By
the Institute of Immunology, Genetics and Metabolism (INIGEM), National Research Council, Argentina
the Division of Periodontics, Section of Oral, Diagnostic and Rehabilitation Sciences, College of Dental Medicine, Columbia University