



# Effect of Temperature on the Dental Implant Osseointegration Development in Low-Density Bone: An *In Vivo* Histological Evaluation

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Implant drilling procedures could cause a temperature rise in the surrounding hard tissue.<sup>1</sup> Many *in vitro* evaluations have demonstrated heat generation and subsequent bone overheating when incorrect surgical protocols were performed.<sup>2,3</sup> Different temperature values were reported in different published articles because a large number of factors are involved during drilling heat generation.<sup>4-6</sup> Pressure applied, drill speed, drill material, drilling depth, drill sharpness, drill diameter, number of drill uses, and irrigation method could significantly influence the temperature rise.<sup>7,8</sup> Albrektsson et al<sup>9</sup> reported that a temperature rise until dangerous level could lead to cell necrosis and degeneration of some proteins. The temperature threshold level for thermal bone tissue injury was first set at 56°C<sup>10</sup> and then Eriksson and Albrektsson's study<sup>11</sup> demonstrated that heating the bone up to 47°C or 50°C for 1 minute could be sufficient to impair the bone formation in rabbit cortical bone. Their study was

**Objectives:** To make an *in vivo* evaluation of the effects of 2 different bone temperatures, on the development of implant osseointegration, in low-density bone.

**Materials and Methods:** Fifteen implant osteotomic sites were prepared in the iliac crests of sheep. Before the implant insertion, 5 sites were heated to 50°C for 1 minute, 5 sites to 60°C for 1 minute, and 5 sites were not overheated. Fifteen titanium dental implants (Cortex, Israel) were inserted. After a healing period of 2 months, the histomorphometric parameters calculated for each implant were the Bone–Implant Contact percentage (%BIC) and the infrabony pocket depth. Unpaired *t* test was applied to find statistical differences between groups.

**Results:** No implants failed. Statistical significant differences in %BIC and periimplant bone loss were found between the 60°C group and control group. No significant differences were found between the 50°C group and control group, although bone suffering signs were present.

**Conclusion:** An osteotomic site overheating up to 60°C for 1 minute in low-density bone, before implant insertion, did not lead to implant failure, but it induced significant crestal bone loss during healing and lower %BIC. (Implant Dent 2015;24:96–100)

**Key Words:** low-density bone, peri-implant bone resorption, bone temperature, osseointegration

performed using a dividable titanium structure with a 1-mm-wide transverse canal in which the authors could evaluate the thorough bone growth. The authors were only able to evaluate the bone growth inside the canal and did not focus on the implant osseointegration because they did not place real titanium dental implants. The titanium structure was inserted in rabbit tibia that is mainly composed of cortical bone, and it is not clarified if the cancellous bone could have the same clinical behavior. A recent study<sup>12</sup> demonstrated histologically

in cortical bone, with an *in vivo* model, that bone overheating due to insufficient bur irrigation, before implant insertion, could have some negative effects on titanium dental implant osseointegration but they did not measure the bone temperature achieved. In a subsequent study, Trisi et al<sup>13</sup> reported that cortical bone heating before implant insertion (sheep mandible) up to 60°C for 1 minute did not impair the osseointegration, but it was enough to create periimplant infrabony pockets statistically deeper than the control group. The

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clinical response of low-density bone heating is still unclear because no study, to our knowledge, evaluated if heating *in vivo* could influence dental implant osseointegration process in cancellous bone. In spongy bone, there are more blood vessels and less trabeculae, and these differences could affect the reaction to overheating. Some authors<sup>14,15</sup> have proposed surgical techniques to minimize the heat generation in cancellous bone during drilling and implant placement procedures as the use of wider diameter burs, low drill speed, or bone condensing technique. The authors of this study want to evaluate the effect of heat stress on dental implant osseointegration in a region, the sheep iliac crest, composed by cancellous bone with a density of 20% to 30%. The objective of this study was to histologically evaluate the effects on the osseointegration of 2 different bone temperatures before dental implant insertion in cancellous bone *in vivo*.

## MATERIALS AND METHODS

The protocol for this study was submitted and approved by the Animal Ethical Committee at the Veterinary School of the University of Teramo (Teramo, Italy).

Two female sheep aged 4 to 5 years old were randomly selected. Exclusion criteria were general contraindications (pregnancy, systemic disease) to implant surgery and active infection or severe inflammation in the area intended for implant placement. The animals were given thiopental sodium (Pentothal, Hoechst, Austria) for induction of anesthesia as needed. After orotracheal intubation and ventilation, anesthesia was sustained with nitrous oxide and oxygen with 0.5% halothane. Physiologic saline was administered for fluid replacement.

The iliac crests were exposed through a skin incision of 15 cm in length. The skin and facial layers were opened and closed separately. After dissection of the soft tissues exposing the bone edge, 3 or 4 implant sites (for a total of 15) were prepared in each (left and right) side of the iliac crest following the recommended manufacturer's drill sequence (the drilling speed was

1000 rpm) under saline solution irrigation. The 15 osteotomic sites were randomly treated in 3 different ways before inserting the implants. In the 50°C group, the osteotomic sites were overheated up to 50°C for 1 minute, whereas in the 60°C group, they were overheated up to 60°C for 1 minute. Control sites were not heated. An electronically controlled device composed by a metal cylindrical probe, 3 mm in diameter and 10 mm in length, connected to a thermostat was used to apply the set temperature stimulation. Every 5 seconds, the hot probe was removed (for 20 seconds) and then was reinserted for 5 seconds again because after 5 seconds the temperature of the probe decreased by 20°C. For this reason, we waited for the temperature of the probe to rise up again to the set temperature measured with a thermocouple. Temperature within the osteotomic site was measured after probe reinsertion to check that the bone temperature was the selected one. This procedure was repeated 12 times for each site for a total of 60 seconds of permanence of the hot probe. All animals received all the 3 treatments. Treatments were randomized to each animal. In a separate register, position data were reported (animal number, iliac crest side, and mesial or distal position) to recognize implant group pertain. Fifteen titanium dental implants with rough surface (Dynamix; Cortex, Shlomi, Israel), 10 mm in length and 3.8 mm in diameter, were positioned. Cover screws were placed over the heads of the fixture.

Surgical wounds were closed by a resorbable periosteal muscular inner suture followed by a cutaneous silk 2-0 external suture. Each animal underwent an antibiotic systemic therapy for 5 days with 8 mL long-acting Clamoxil (Pfizer Limited, Sandwich, MA). The sheep were killed 2 months after implantation by an overdose of pentothal sodium (Thiopental).

The specimens were immediately fixed in 10% neutral buffered formalin. After dehydration, the specimens were infiltrated with a methyl methacrylate resin from a starting solution 50% ethanol/resin and subsequently 100% resin, with each step lasting 24 hours. After polymerization, the blocks were

**Table 1.** Average Values of %BIC of Every Group After 2 Months in Soft Bone (Sheep Iliac Crest)

	Average %BIC $\pm$ SD
Control group	38.05 $\pm$ 1.384
50°C group	25.92 $\pm$ 5.424
60°C group	25.42 $\pm$ 1.492

The control group showed the highest value of %BIC between groups.

sectioned and then ground down to about 40  $\mu$ m. Toluidine blue staining was used to analyze the different ages and remodeling pattern of the bone. Histomorphometric analysis was performed by digitizing the images from the microscope by means of a JVC TK-C1380 Color Video Camera (JVC Victor Company, Yokohama, Japan) and a frame grabber. The images were acquired with a 10 $\times$  objective including the entire implant surface. Subsequently, the digitized images were analyzed by the image analysis software IAS 2000 (Delta Sistemi, Roma, Italy). For each section, the 2 most central sections were analyzed and morphometrically measured. The histomorphometric parameters calculated using the software were the Bone–Implant Contact percentage (%BIC), which represents the linear surface of the implant directly contacted by the mineralized bone matrix, and the infrabony pocket depth. An unpaired *t* test was applied to test the statistical differences between the different groups using the statistical software GraphPad Prism 5 (www.graphpad.com).

## RESULTS

No implant failure was registered after 2 months of healing.

**Table 2.** Statistical Comparison (Unpaired *t* Test) of Average %BIC Value Between Groups in the Soft Bone

%BIC Comparison	<i>P</i>
Control vs 50°C group	0.092*
Control vs 60°C group	0.0003†
50°C vs 60°C group	0.931*

The 60°C group showed a statistically lower %BIC than the control group.

\*Significant.

†Nonsignificant.



**Table 3.** Average of Infrabony Pocket Depth in Each Groups

	Average Infrabony Pockets Depth $\pm$ SD (mm)
Control group	1.212 $\pm$ 0.1570
50°C group	3.028 $\pm$ 0.8095
60°C group	3.110 $\pm$ 0.3283

In the control group, the average infrabony pocket depth was less than half of the one in the heated sites group.

#### Histomorphometric Analysis

The average %BIC values are reported in the Table 1. The statistical comparison of %BIC values using unpaired *t* test revealed statistical significant differences only between the 60°C and control groups (Table 2). The average infrabony pocket depths for each group are reported in Table 3. Significant differences were found in infrabony pocket depth between implants of the 60°C and control groups (Table 4). Implants in the 50°C group showed signs of bone suffering (lower %BIC and crestal bone loss) more pronounced with respect to the control group, but these differences were not statistically significant.

#### Histological Evaluation

Implants of all groups showed achievement of osseointegration in the apical area with newly formed bone integrated on the titanium surface. Some areas of lamellar bone in direct contact to implant threads were also found. Areas of direct bone contact alternated with areas of bone marrow along the implant interface. Bands of osteoid tissue coupled with areas of bone resorption were observed testifying a normal remodeling pattern. Signs

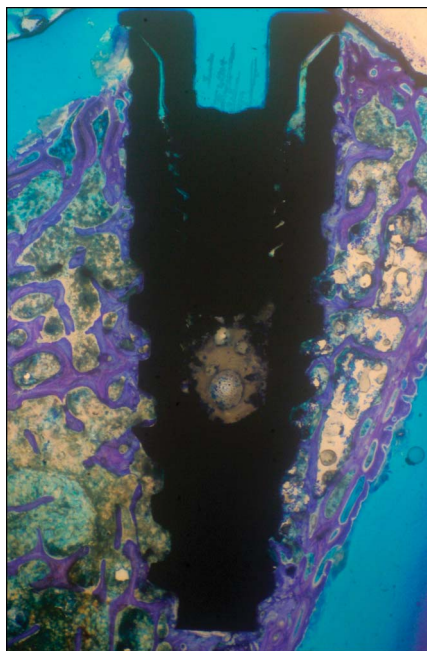
**Table 4.** Statistical Comparison (Unpaired *t* Test) of Infrabony Pocket Depth Average Between Groups

Infrabony Pockets Depth Average Comparison	<i>P</i>
Control vs 50°C group	0.092*
Control vs 60°C group	0.003†
50°C vs 60° group	0.927*

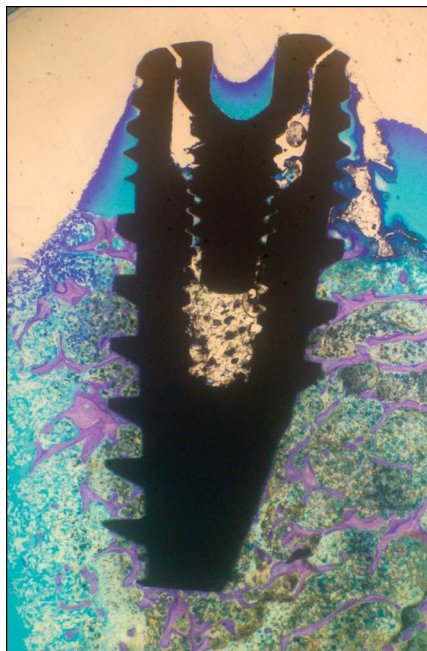
The 60°C group showed infrabony pockets statistically deeper than the control group.

\*Significant.

†Nonsignificant.



**Fig. 1.** Representative implant in the control group. Implants achieve osseointegration with new bone formation in direct contact to the titanium surface.



**Fig. 2.** Representative implant in the 50°C group. Implants showed some signs of bone resorption.

of bone remodeling and absence of inflammatory processes were evident in the control group (Fig. 1). Implants in the 50°C group healed with some crestal bone loss (Fig. 2). Inflammatory



**Fig. 3.** Representative implant in the 60°C group. Crestal bone resorption was evident.

cells were not present, and bone loss was replaced by dense connective tissue. Implants in the 60°C group showed diffused infrabony pockets and significant crestal bone resorption (Fig. 3). A huge inflammatory process surrounding the implants in this group was evident. The connective tissue was rich in vessels and was infiltrated by inflammatory cells such as plasma cells, macrophages, and polymorphonuclear cells.

#### DISCUSSION

The atraumatic surgical technique avoiding overheating of the implant site and the primary stability are reported as 2 of the main prerequisites involved in a successful osseointegration development.<sup>16–20</sup> Eriksson and Albrektsson<sup>11</sup> established the temperature threshold level of bone injury at 47°C or 50°C for 1 minute. They showed, using a titanium structure named Bone Growth Chamber inserted in rabbit tibia, that a temperature of 47°C for 1 minute significantly impaired the bone growth through the canal of the chamber compared with the control group. The temperature was measured at 0.5 mm from the titanium structure and the real temperature at the interface bone titanium could be underestimated

probably. They reported that an 8°C higher temperature was measured directly on the titanium structure surface compared with that measured at 0.5 mm. The authors did not test the effect of these temperature levels on dental implant osseointegration development and did not consider the effects of different bone densities. An old study by Lundsdog<sup>21</sup>, which was in contrast with Eriksson and Albrektsson's<sup>22</sup> data, reported that the temperature threshold level necessary to prevent bone regeneration was 70°C for 30 seconds. In this study, temperatures up to 50°C for 1 minute in low-density bone did not impair significantly the dental titanium implant osseointegration, whereas a bone temperature of 60°C for 1 minute seemed to influence negatively the %BIC after 2 months of healing moreover. Crestal bone resorption around dental implants was also significantly higher in the 60°C group with respect to other groups. Implants in the 50°C group showed bone resorption and %BIC values lower than control group, but without statistical significance. Temperature effect on implant osseointegration in cortical bone, 50°C and 60°C for 1 minute, were tested *in vivo* in a recent study.<sup>13</sup> This study showed no statistical differences in %BIC between the groups, and only in the 60°C group 1 was reported to induce a significant periimplant crestal bone resorption. In this study, the effect of temperature on osseointegration, using these temperature levels, were analyzed in the cancellous bone. As explained before, our results suggest that a bone temperature of 60°C for 1 minute in low-density spongy bone induces significant damaging to the osseointegration process. The cancellous bone is probably less prone than the cortical bone to overheating during drilling procedure,<sup>23</sup> but the heat stress, applied before dental implant insertion, was able to impair the osseointegration development. Comparing the results of this study with the same type of evaluation on cortical bone (Trisi et al., 2013, in press), it is evident that low-density bone seems to be more frail to heat-induced damage than high-density bone. The reason for these results could be explained by the different structure

of the 2 bones. It is important to focus the attention on the requisites for a correct bone healing: an adequate number of cells, blood supply, and functional stimulus. The bone tissue is dependent on an adequate supply of nutrient, transported through the vascular system. When the bone temperature rose up beyond physiological levels, an immediate vascular reaction and abnormal changing in the fat cells were demonstrated.<sup>24,25</sup> The cortical bone has a compact structure and minimal gaps within the inorganic portion. The main blood supply of cortical bone (high-density bone) is given by periosteal vessels, and only a few small vessels flow into the lamellar structure (Haversian and Volkmann canals). The heat-induced damage on cortical bone did not impair, probably, the blood supply because the periosteum, which contains the main vessels, is not damaged.<sup>26</sup> The low-density bone is mainly composed of bone trabeculae filled by a network of rod- and plate-like elements and contains a great number of blood vessels and bone marrow. The blood supply in the cancellous bone originates from the marrow vessel. When the same heat stress is given to low-density bone, the blood vessels and the bone marrow are damaged and the consequent transport of nourishment to the bone cells is impaired, with bone resorption as a consequence. Eichler and Berg<sup>27</sup> considered the blood flow to have a significant influence on the extent of the thermal injury and the study by Krause et al<sup>28</sup> also stated that the blood flow has a significance only in cancellous bone. The heat sensibility of the blood vessels seems to be of crucial importance, and the different clinical behavior to heat stress between cortical and cancellous bone could be explained with their different structures and different blood supply systems. Furthermore, the heat impact may be combined with other bone injuries as the ischemia. The ischemia due to irreversible damage to marrow vessels and intercellular substance could reduce also the regeneration properties of cancellous bone after the overheating trauma. The implant osseointegration in low-density bone, rich in intercellular substance, vessels, and cells, was significantly impaired after heating

the site up to 60°C for 1 minute. Other studies from the literature<sup>29,30</sup> indicate temperatures above 53°C responsible for damage of the bone intercellular substance. Additional studies are needed to confirm the results of this study.

## CONCLUSION

Dental implant osseointegration in low-density bone is susceptible to heat-induced damage. Results of this study demonstrated that a bone temperature of 60°C for 1 minute is sufficient to significantly reduce the implant %BIC. Periimplant crestal loss observed both in the 50°C and 60°C groups indicates bone suffering due to overheating. A temperature of 50°C for 1 minute, in low-density bone, may impair the osseointegration development with an unpredictable crestal bone resorption. For 1 minute, 60°C seems to be sufficient to significantly reduce the implant osseointegration percentage.

From a comparison of these results with those of a same study on cortical bone (Trisi et al., 2013, in press) appears that osseointegration in low-density bone is more subject to heat-induced injury. It is important to avoid implant surgical procedure that could lead to overheating of the bone matrix before implant insertion.

## DISCLOSURE

The authors declare that they have no financial relationship or interest that may pose a conflict of interest between them and Cortex Dental Implants (Shlomi, Israel).

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