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Investigation of maxillary sinus bone graft healing by MicroCT

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ABSTRACT

Objective: A MicroCT analysis of bone healing quality and quantity subsequent to placement of a reformulated tricalcium phosphate (TCP)* in maxillary sinus ridge augmentation. Methods: Ten (10) trephined rod shaped human bone cores were obtained from an independent source at three months after the placement of tricalcium phosphate particulate in the maxillary sinus for ridge augmentation prior to implant placement. The samples were immediately stored in 10% neutral buffered formalin. Using SCANCO 40 MicroCT machine, the samples were evaluated at resolutions of 6 and 20µm respectively. The threshold for bone and graft material was determined using visual image, intensity (grey level) and histogram analyses. The density and three dimensional micro architecture of the bone and graft material were analyzed using the software which was interactive with the investigators. Results: The samples were investigated for micro architecture and density of the bone and graft material. Along with mineral density, the trabecular bone parameters such as Trabecular thickness (Tb Th), Trabecular separation (Tb sp), Structural Model Index (SMI), Trabecular number (Tb no), and connectivity density were obtained for each sample and analyzed ANOVA ($p \leq 0.05$). The results obtained at both resolutions (6and 20µm) were comparable, however parameters obtained at 20µm will be used in the future studies including histological, histomorphometrical and scanning electron microscopy comparisons. Conclusion: MicroCT allowed the study of bone both quantitatively and qualitatively without destroying the samples. These results demonstrated an osteoconductive effect of TCP. The mean bone and graft volume obtained was 25.5 and .43 mm3 respectively; suggesting new bone formation and graft dissolution. These results corroborate independent reports of implants having moved to functional status without failures. The data achieved can be further used to correlate with the values obtained from the traditional methods.

INTRODUCTION

Evaluation of regenerated bone subsequent to graft placement has traditionally been analyzed by histological and histomorphometric methods. These employ two dimensional histological sections. In addition to being tedious and destructive, they base many of their estimations of three dimensional anatomies on simplified models of trabecular bone shapes.(Singh, 1978) Further, the analysis of a few sections cannot be fully representative of an entire sample. Serial sectioning is also done but that is often too destructive and labor intensive(Odgaard *et al.*, 1994).

Newer imaging techniques have made it easier to obtain the high resolution three dimensional images necessary to examine the bone architecture directly(Feldkamp *et al.*, 1989); The quantity and quality includes not only the bone volume and density but also the parameters that describe the microarchitecture of the bone formed. This is important since in addition to bone mineral density, the three dimensional bone microarchitecture strongly influences the mechanical properties of the cancellous bone(Borah *et al.*, 2000; Jensen *et al.*, 1990; Odgaard, 1997; Siffert *et al.*, 1996; Ulrich *et al.*, 1999). It has also been shown that the the MicroCT analysis of bone can be correlated with the values obtained from the traditional histological and histomorphometric methods.(Fajardo and Muller, 2001; Muller *et al.*, 1996; Muller *et al.*, 1998)

In this pilot study we analyzed the regenerated bone structure subsequent to placement of graft material, (tricalcium phosphate, TCP) in the maxillary sinus ridge augmentation using Micro-computed tomographic analysis.

MATERIALS AND METHODS

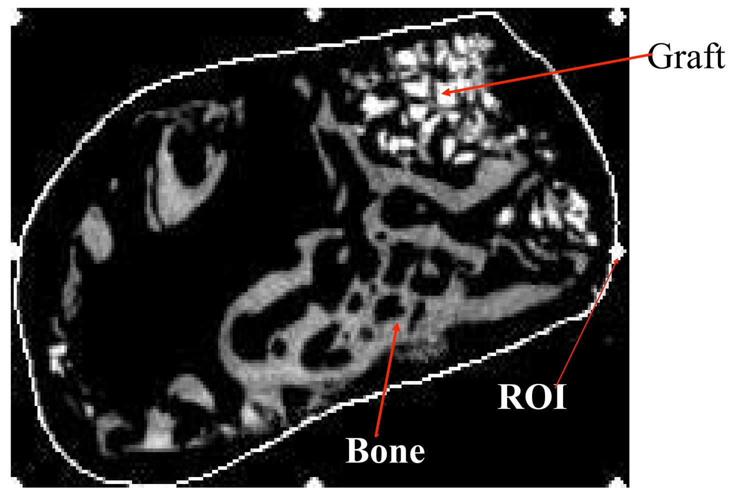
Sample: Ten rod shaped human bone cores obtained by trephine, three months after the placement of FDA approved synthetic tricalcium phosphate bone graft material in the maxillary sinus for ridge augmentation prior to implant placement. The samples were immediately stored in 10% neutral buffered formalin (Figure 1)

Machine: A high resolution, desktop Microtomographic imaging system (µCT40.scanco Medical AG, Bassersdorf, Switzerland) (Figure 2).

Human Bone cores
3 -9 mm (L), 4mm (D)

Sample Loading

Desktop Microtomographic imaging system (µCT40.scanco Medical AG, Bassersdorf, Switzerland)

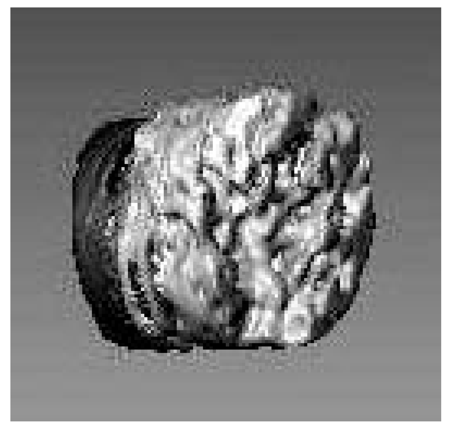


2-D Horizontal section/ slice/ tomograph at 20µm

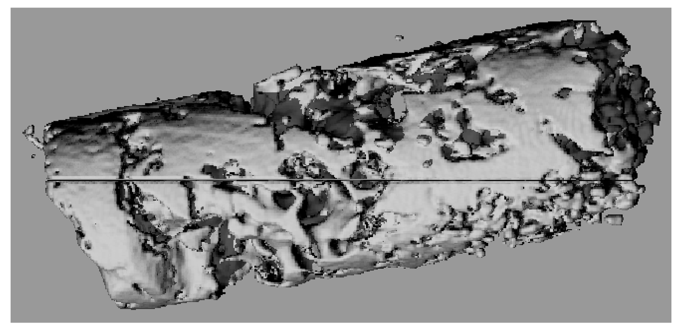
A pilot study using 37 µm resolution gave inconclusive results leading to the above mentioned machine where the biopsies were scanned at 6 and 20 µm resolutions. The samples were snugly fit in the sample holder sized 10 mm along with formalin. They were scanned at 70KeV with each voxel size being 6 and 20µm for respective resolutions. The numbers of slices per sample were variable; up to 1000 slices were obtained depending on the size of the sample. The orientation of the sample when scanning was horizontal since the 2-D tomographs then obtained could be directly used for comparison with histological sections. The Region of Interest (ROI), which in this case, was the entire slice containing the TCP and the bone was defined followed by thresholding for the powder and bone at 235 (grey level magnitude) and only powder at 600 (grey level magnitude). As a control, TCP was scanned separately, however it was concluded that the threshold then obtained could not be used directly in the study as the combination of bone and graft was unique and TCP powder did not emulate the TCP in this combination. The volume, density and three dimensional micro architecture of both the bone and graft material were analyzed using the software which was interactive with the investigators

RESULTS

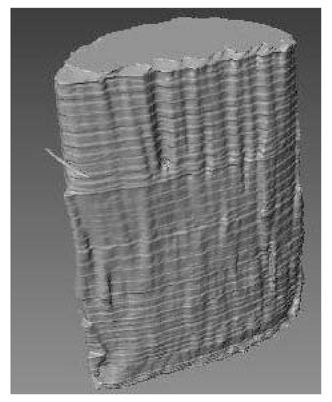
Results at 37µm were inconclusive and differentiating between powder and bone was not possible .



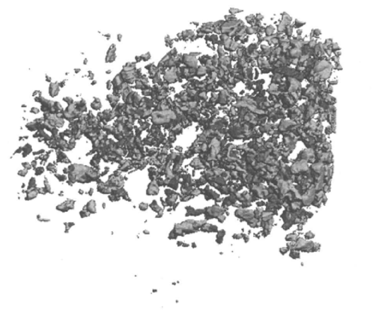
3-D reconstruction at 37µm



3-D reconstruction at 6µm



Graft material at 37µm



Graft material at 6µm

The density, volume and three dimensional micro architecture of both the bone and graft material was computed at 6 and 20 µm. Micro architecture of the trabecular bone was described by parameters such as trabecular thickness (Tb Th), trabecular separation (Tb sp), structural model index (SMI), trabecular number (Tb no), and connectivity density. The results obtained at 6 and 20 µm were compared using a Student's T-Test analysis. The data obtained are shown below.

Variable	Resolution (µm) Mean, (SD)		p-value < .05
	6µm	20µm	
Graft Volume	2.25 (1.83)	.42 (.37)	**0.0120**
Bone Volume	23.77 (8.46)	25.50 (11.28)	0.7019
Graft Density	2069.2 (31.82)	1992.3 (28.56)	**0.0001**
Bone Density	1076.4 (80.87)	1056.3 (81.99)	0.5871
Total Volume	26.02 (9.85)	20.93 (11.57)	0.9853

Table 1: T-Test analysis showing significant difference in Graft volume, density at 6 and 20 µm.

CONCLUSIONS

The current MicroCT machine can be used to evaluate healing bone characteristics in the present study.
A significant difference in graft volume and density at 6 and 20 µm were found.
The bone volume was more than the graft volume at both 6 and 20 µm.
The bone density was less than graft density at both 6 and 20 µm.
These results can further be compared and analyzed with histomorphometric and histological data.

REFERENCES

Borah B, Dufresne TE, Cockman MD, Gross GJ, Sod EW, Myers WR, Combs KS, Higgins RE, Pierce SA, Stevens ML (2000). Evaluation of changes in trabecular bone architecture and mechanical properties of minipig vertebrae by three-dimensional magnetic resonance microimaging and finite element modeling. *J Bone Miner Res* 15(9):1786-97.
Fajardo RJ, Muller R (2001). Three-dimensional analysis of nonhuman primate trabecular architecture using micro-computed tomography. *Am J Phys Anthropol* 115(4):327-36.
Feldkamp LA, Goldstein SA, Parfitt AM, Jesion G, Kleerekoper M (1989). The direct examination of three-dimensional bone architecture in vitro by computed tomography. *J Bone Miner Res* 4(1):3-11.
Jensen KS, Mosekilde L, Mosekilde L (1990). A model of vertebral trabecular bone architecture and its mechanical properties. *Bone* 11(6): 417-23.
Muller R, Hahn M, Vogel M, Delling G, Ruegsegger P (1996). Morphometric analysis of noninvasively assessed bone biopsies: comparison of high-resolution computed tomography and histologic sections. *Bone* 18(3):215-20.
Muller R, Van Campenhout H, Van Damme B, Van Der Perre G, Dequeker J, Hildebrand T, Ruegsegger P (1998). Morphometric analysis of human bone biopsies: a quantitative structural comparison of histological sections and micro-computed tomography. *Bone* 23(1):59-66.
Odgaard A, Andersen K, Ullerup R, Frich LH, Melsen F (1994). Three-dimensional reconstruction of entire vertebral bodies. *Bone* 15(3): 335-42.
Odgaard A (1997). Three-dimensional methods for quantification of cancellous bone architecture. *Bone* 20(4):315-28.
Siffert RS, Luo GM, Cowin SC, Kaufman JJ (1996). Dynamic relationships of trabecular bone density, architecture, and strength in a computational model of osteopenia. *Bone* 18(2):197-206.
Singh I (1978). The architecture of cancellous bone. *J Anat* 127(Pt 2):305-10.
Ulrich D, van Rietbergen B, Laib A, Ruegsegger P (1999). The ability of three-dimensional structural indices to reflect mechanical aspects of trabecular bone. *Bone* 25(1):55-60.

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