Porous Hydroxyapatite for Grafting the Maxillary Sinus in Sheep: Comparative Pullout Study of Dental Implants

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Purpose: *In this experimental study, dental implants placed after maxillary sinus grafting with either porous hydroxyapatite (HA) (Interpore 200) or autogenous bone were examined for their mechanical stress tolerance.* ***Materials and Methods:*** *A total of 54 titanium plasma flame–sprayed cylindric implants were placed in the lateral sinus wall bilaterally of 27 mountain sheep. The bony sinuses were opened through an extraoral approach. Eighteen sinuses were grafted with porous HA, and another 18 were grafted with cancellous bone from the iliac crest. Eighteen non-grafted sinuses were used as controls. In the same operation, 2 cylindric implants were placed in each of the sinuses. One implant of each sinus was tested for mechanical strength of the bone-implant interface at 12, 16, and 26 weeks using pullout force.* ***Results:*** *The mean pullout force was 259.3 N in the control group, 356.7 N in the autogenous bone group, and 376 N in the HA group. Pooled data for the grafted sites showed the pullout force to be significantly higher than in the empty control sites (P = .02). The pullout force increased significantly with ongoing healing time (P = .02), but there was considerable variation within the groups. While the force remained more or less constant throughout the follow-up time in the controls (248 N at week 12 to 276 N at week 26), it increased dramatically in the group augmented with autogenous bone (223.8 N at week 12 to 523.16 N at week 26). The pullout force was initially highest in the HA group (302.3 N at week 12) and increased to 423.5 N at 26 weeks, but it did not reach the levels recorded in the autografted group.* ***Discussion and Conclusion:*** *Mechanical tests of bone-to-implant contact in a sheep model showed that HA for 1-stage sinus floor elevation significantly increased the pullout force versus ungrafted sinuses, although it was less than that found with autogenous bone after 26 weeks.* INT J ORAL MAXILLOFAC IMPLANTS 2003;18:691–696

Key words: *dental implants, hydroxyapatites, pullout force, sinus augmentation*

Because of its interconnected, 3-dimensional pore system, with a pore size of about 190 to 230 nm and a particle size of about 425 to 1,000 nm, its biocompatibility, and non-toxicity, coralline hydroxyapatite (HA) possesses properties that are important for a useful osteoconductive bone substitute.[1] The ingrowth of bone and connective tissue into the porous meshwork is thought to provide early implant stability.[2] The pore system of HA serves as a scaffold for blood vessels, has a beneficial effect on osteoneogenesis,[3] and contributes to an increased number of bone-to-implant contacts.[4] HA has been successfully used for sinus grafting in several clinical[5] and experimental studies,[4,6] which have suggested that it is a suitable bone substitute capable of improving the quality of the implant host site.[3] To the authors' knowledge, the biomechanical strength of dental implants placed after grafting the maxillary host bone with porous HA has not been investigated.

In an experimental study, the bone-to-implant contact following sinus floor elevation with HA or autogenous bone was examined histomorphometrically in 27 adult female mountain sheep and compared with an ungrafted control group.[7] The groups

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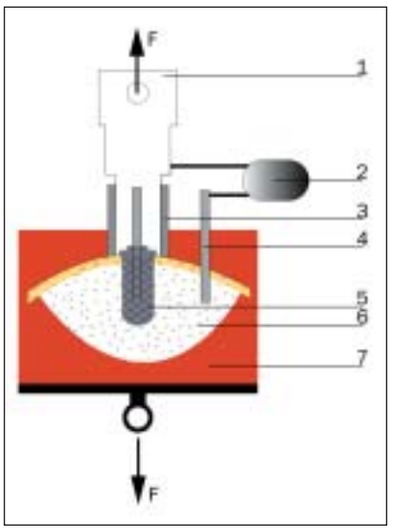

Fig 1 *(Left)* Titanium plasma flame–sprayed cylindric implant.

Fig 2 *(Right)* Schematic of a mechanical test implant. F = force; 1 = extended yoke; 2 = extensometer; 3 = plastic sleeve; 4 = retaining screw; 5 = implant; 6 = bone with augmentation material; 7 = embedding material (polymethyl methacrylate resin).

grafted with porous HA (P = .002) and cancellous bone from the iliac crest (P = .0005) showed significantly more bone apposition at the implant surface than the controls. These histologic and histomorphometric data suggest that, together with other properties, the mechanical strength of HA makes it a promising candidate for a bone substitute.

The present study was designed to establish whether the histologically documented increased bone apposition after sinus floor elevation with HA is associated with improved implant stability.

MATERIALS AND METHODS

The study design and the surgical procedure have been described elsewhere.[8] Two rotationally symmetric, cylindric, titanium plasma flame–sprayed implants (Friatec, Friedrichsfeld, Germany) with a diameter of 3.75 mm and a length of 8 mm were placed in each maxillary sinus of 27 adult mountain sheep (mean body weight, 60.8 ± 7.4 kg). Their surface showed a flat, checkered groove pattern (Fig 1). Through an extraoral approach, a 1×1-cm window was cut into the buccal bony sinus wall. The subjacent membrane was carefully elevated to create space on the sinus floor for the placement of 2 implants distal to the osteotomy site. Of the 54 sinuses, 18 were packed with porous HA (Interpore 200, Interpore International, Irvine, CA); another 18 were packed with cancellous bone from the iliac crest. The remaining 18 sinuses were implanted without grafting and served as controls. The osteotomy defects were covered with soft tissue only. All animals were given penicillin (2 doses of 10 mIU units of Penicillin G Sodium; Hoechst, Vienna, Austria) and oxacillin (1 g Stapenor; Bayer, Leverkusen, Germany) for 3 days postsurgery. The implants were left covered throughout the follow-up time.

Specimen Preparation

At 12, 16, and 26 weeks post-implantation, 3 groups of 9 animals each were sacrificed by an overdose of thiopental (Tyrol Pharma, Vienna, Austria) and embutramide (T61; Hoechst, Vienna, Austria). Six sinuses with 2 implants each were available for evaluation in each treatment group at each sampling time. They were randomly allocated to histologic and mechanical analysis.[9]

The block specimens selected for mechanical testing were stored in buffered formalin solution until further use. Excess bone was removed with a band saw (Exakt Apparatebau, Norderstedt, Germany). The cover screws were removed and replaced by flushly fitting retaining screws anchored in the inferior implant threads. These transmitted the force to the implants. A plastic sleeve with an interior diameter of 4.5 mm and a length of 15 to 20 mm was placed around the threads of the screws. This sleeve guaranteed interference-free gliding of the screws and implants in the resin during mechanical testing by keeping off the embedding material (Fig 2). To ensure that the pullout direction coincided with the implant axis, the samples were clamped down in the testing unit (Universal Testing Machine RM 250; Schenck, Munich, Germany) with the retaining screw during embedding in autopolymerizing polymethyl methacrylate resin (modulus of elasticity, 3,353 ± 235 N/mm^2) (Technovit 4071, Kulzer, Wehrheim, Germany). Implant

Table 1 Mean Pullout Force (Least-Square Means ± SEM) of the Implants of 3 Different Groups for Each Observation Period

Group	12 weeks	16 weeks	26 weeks	Overall
Control	248.0 ± 99.90	260.2 ± 180.10	269.8 ± 50.40	259.3 ± 168.40
Cancellous bone	223.8[a] ± 127.59	323.2 ± 105.83	523.7 ± 200.26	356.7[b] ± 144.60
Hydroxyapatite	301.3 ± 116.40	402.2 ± 199.60	423.5 ± 164.40	376.0 ± 163.10

[a]$P < .05$ vs cancellous bone at 26 weeks; [b]$P < .05$ vs negative control group at 12 weeks.

parts possibly penetrating the Schneiderian membrane were covered with a plastic material before embedding to prevent direct contact between the embedding material and the implants. A curing time of at least 30 minutes was observed. During polymerization, the testing unit was operated in a force-controlled range of less than 8 N.

For testing, the pullout force was increased from 0 to 1,000 N at a speed of 0.4 mm/min until the bone-to-implant contact failed. The maximum pullout force was determined from stress-strain diagrams.

Statistical Analysis

Analysis of variance was used to assess possible differences in pullout force between the groups (cancellous bone, HA, and controls) and the different observation periods (12, 16, and 26 weeks). Possible interactions between the 3 groups and time were also evaluated. All P values were 2-sided; $P < .05$ was considered statistically significant. Multiple paired comparisons were adjusted according to Tukey-Kramer.

RESULTS

All animals tolerated the surgery well. The postoperative course was uneventful. Two implants of the autogenous bone group retrieved at 26 weeks were excluded from analysis. One was no longer in situ at the time of sampling; the other was discarded because of a processing mistake made during specimen preparation.

Mean pullout forces throughout the entire follow-up time were 259.3 N for the controls, 356.7 N for the autogenous bone group, and 376.0 N for the HA group. Pooled pullout forces of the grafted groups were significantly higher than those in the controls ($P = .02$). The differences between the grafted groups were borderline significant ($P = .069$) (Table 1).

Mean pullout forces of all implants were evaluated in a group analysis (Table 2). They were 258.04 N at 12 weeks, 328.52 N at 16 weeks, and 404.56 N at 26 weeks. Time was shown to have a significant effect ($P = .02$) (Table 1); pullout strengths at 16 weeks ($P < .0001$) and 26 weeks ($P < .0001$) were significantly higher than at 12 weeks. Generally, the time effect tended to be linear ($P = .006$).

Table 2 Analysis of Variance for the Total Model, Described by the Sum of Squares, the F value, and the *P* value and Split to Covariates of the Model in Type II Sum of Squares

	df	SS	F value	*P* value
Model	8	447512.7	2.62	.02
Week	2	183420.4	4.30	.02
Type	2	121498.2	2.85	.07
Type · week	4	127382.2	1.49	.22
Error	41	873784.9		
Corrected total	49	1321297.6		

df = degree of freedom; SS = sum of squares; F value = Fisher value.

The increase in pullout force varied among the groups tested (Fig 3). Assuming the time course to be linear, the increase in the control group was slight (1.6 N/week), but it was relatively constant and clearly lower after 16 weeks than in the other groups. The group augmented with autogenous bone showed the lowest pullout forces at 12 weeks, but these steadily increased by 21.4 N/week to levels unmatched by any other group at week 26. In the HA group, pullout forces were highest initially but followed a pattern clearly different from the steady rise seen in the other groups. Pullout force increased by 25.2 N/week between weeks 12 and 16 and by 2.13 N between weeks 16 and 26. This was equivalent to a weekly gain of 8.7 N throughout the follow-up time (weeks 12 to 26) (Table 3, Fig 3).

DISCUSSION

In this experimental study, the strength of the bone-to-implant contact following sinus grafting with porous HA or autogenous bone was investigated in sheep. For this purpose, the implants were subjected to pullout tests at 12, 16, and 26 weeks.

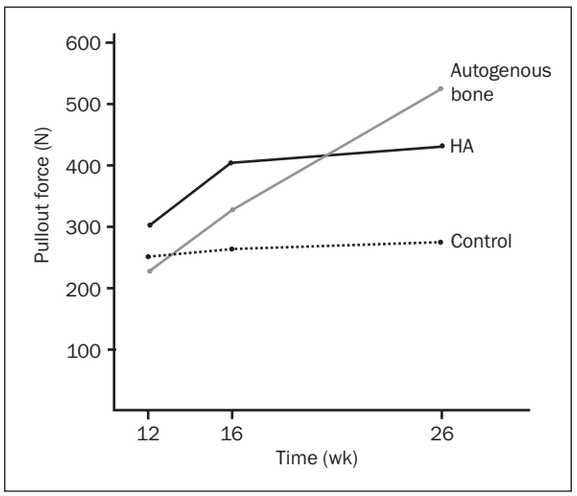

Table 3 Weekly Increase in Pullout Force (N) During the Observation Period

	Time period (wk)		
Group	**12–16**	**16–26**	**12–26**
Control	3.05	0.96	1.56
Cancellous bone	24.85	20.05	21.42
Hydroxyapatite	25.23	2.13	8.73

Fig 3 *(Left)* Time-dependent increase in pullout force in the 3 different groups (HA, bone, ungrafted controls).

The use of mechanical tests such as pullout or pushout tests for evaluating the interfacial shear strength of bone-to-implant contact has been controversial. Using numeric stress analysis models, Soltész and Bernauer[10] and Soltész and Baudendistel[11] were able to show that high peak stresses occur at the bone-to-implant interface, resulting in premature failure, thus providing an underestimation of the bone-to-implant contact.

Specimen fixation in formalin increases collagen cross-linking and can alter the properties of bone tissue. Therefore, mechanical testing of formalin-fixed samples only provides data relative to other fixed samples, and results obtained from individual samples likely will not provide an accurate measure of the true properties of bone.[12,13]

This study focused especially on the relationship between measuring the results of the individual groups and a comparison of the mechanical data with histologic findings, rather than assessing the absolute strength of the bone-to-implant contact. However, it can be assumed that the bone-to-implant interface can tolerate higher loads than those described in this study.

The pooled data for the 2 grafted groups showed the pullout forces to be significantly higher than those in the controls (P = .02). Despite major between-group differences, their significance was borderline (P = .069). This may be attributable to the relatively wide variability of the test data. Given a larger number of specimens, the differences may well have reached statistical significance.

The mean pullout force was lowest in the controls (259.3 N), irrespective of time. This is roughly equivalent to an anchorage capacity of 294 to 490 N reported by Brånemark for screw-type implants in the dog maxilla.[14] Mean pullout forces in the autogenous bone group were 356.7 N and 375.6 N in the HA group. In a comparable study design, pullout forces with homogenous demineralized freeze-dried bone (365.2 N)[15] and natural bone mineral (393.0 N)[9] were roughly the same.

The implant residence time was shown to have a significant effect on the load tolerance of implants (P = .02). In all groups, the pullout force was found to increase during the follow-up time. This increase tended to be linear over time (P = .006). A time-dependent increase in torque or pullout force was also reported by Tjellstrom and coworkers[16] in human mastoid bone and by Kraut and associates[17] in a goat model. Sennerby and colleagues,[18] by contrast, did not find an increase in the pullout force needed to remove tibial implants from 1.5 to 6 months.

The increase in the pullout forces took a variable course in the groups evaluated. In the empty control group, a relatively slight but constant increase between weeks 12 and 26 was found (from 248 to 269 N). The group augmented with cancellous bone also showed a more or less linear, but more pronounced, increase in pullout force (from 223.8 to 523.7 N). In the HA group, an entirely different time course was seen. As in the autografted group, the pullout force increased dramatically by about 100 N between weeks 12 and 16. After week 16, it increased by no more than 22 N, as can be seen from the flattening of the curve (Fig 3).

The mechanical stress tolerance of other bone substitutes has been dissimilar to that of porous HA for sinus grafting. While initially unchanged, the pullout forces recorded for bovine HA increased dramatically after week 16 to match those of bone autografts (521.8 N at week 26).[9] This was also seen

with homogenous demineralized freeze-dried bone (DFDB) (481.4 N at 26 weeks).[15] With heterogeneous DFDB, by contrast, the pullout force initially decreased, with a subsequent slight increase up to week 26 (325.4 N) without reaching the forces recorded for autogenous bone.[15]

A comparison of the mechanical and the histomorphometric data[7] showed a similar result. Bone-to-implant contacts were comparable in the HA and the autogenous bone group (5.7 ± 0.3 mm for bone autografts versus 5.9 ± 0.3 mm for HA), but they were significantly more extensive than those seen in the controls. The grafted groups did significantly better than the controls, both mechanically (P = .02) and histologically (P = .0002 for HA and P = .0005 for autogenous bone), and time was shown to be a significant factor in both mechanical (P = .02) and histologic evaluation (P = .04).

Nevertheless, the time course of the histomorphometric and mechanical data was not indicative of a direct effect of the bone-to-implant interface on the mechanical stress tolerance in pullout tests. As in the mechanical tests, the percent bone-to-implant contact was lowest in the controls and dropped to about baseline levels at week 26.

In the autogenous bone group, bone-to-implant contact increased steadily, albeit slightly, by about 5%. The dramatic increase in pullout forces between weeks 16 and 26 was, however, not reflected by the histomorphometric data.

In the HA group, bone-to-implant contact clearly decreased between weeks 12 and 16 to increase dramatically after week 16. This sharply contrasts with the pullout tests, which initially showed a major increase between weeks 12 and 16, followed by only slight increases later. The factors underlying this discrepancy are still speculative and should be clarified in further studies. One of them may be that the mechanical stress tolerance of implants is determined not only by the strength of the bone-to-implant interface; neighboring tissues may well be an important contributing factor.

In contrast to these observations, the pullout force and the percent of bone-to-implant contact were reported to be significantly correlated by Chang and coworkers[19] in monkeys and by Kraut and associates[17] in goats. Brånemark and colleagues[20] also reported a statistically significant correlation between pullout force, bone-implant contact length, and peri-implant bone density in the rat tibia, but they surmised that the thickness and density of the local host bone was more important in pullout tests of the screw-type implants tested than the bone-to-implant contact.

When animal data is extrapolated to humans, the differential bone turnover rate in sheep should be given appropriate attention. Szyszkowitz and coworkers reported that fractures in sheep healed 6 to 8 times faster than in humans.[21] Since the implants in this study were left unloaded to ensure comparability of the results, no conclusions can be drawn about a possible change of bone-to-implant contact on mechanically loaded implants.

CONCLUSION

In this study, the biomechanical qualities of porous HA for grafting the maxillary sinus in conjunction with the placement of cylindric dental implants were investigated in sheep. The pullout forces recorded for implants in HA-grafted sinuses were superior to those recorded for implants placed in the unaugmented controls. While they increased significantly over time, they did not match those of autogenous bone at the end of the 26-week follow-up time.

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