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A Comparative Study of Osseointegration of Titanium Implants in Corticocancellous Block and Corticocancellous Chip Grafts in Canine Ilium

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Purpose: This study was undertaken to compare the relative rates and extent of osseointegration of dental implants when placed simultaneously with either corticocancellous block or particulate corticocancellous bone grafts.

Materials and Methods: Using the canine ileum as a model site, the implants were placed so that each served as its own control. The implants were harvested at 1, 2, or 3 months for evaluation by light microscopy, microradiography, and histomorphometry.

Results: Both types of grafts were determined to be viable by microscopic evaluation of fluorescent labels. Qualitatively there appeared to be greater bone density in the corticocancellous block graft implant sites. At 3 months, the block graft implant sites had a level of osseointegration (59.6%) that approximated the control implant sites (65.2%), but was significantly greater than the particulate graft sites (39.2%).

Conclusions: These results indicate that implants in corticocancellous block grafts develop osseointegration more rapidly than those in particulate bone grafts. The clinical implications of these findings are discussed.

The reconstruction of the severely atrophic jaw has challenged the ingenuity of the reconstructive surgeon. Among the substances used to augment and maintain the bony mass of the jaws have been autogenous corticocancellous bone in both block and particulate forms. The ability to maintain ridge mass has, however, been disappointing. Endosseous implants in conjunction with onlayed autogenous bone grafts have been successfully used in the treatment of the edentulous jaw. Simultaneous grafting and implant placement has also proved to be a successful procedure to maintain implant health, and bony integrity and mass. This procedure was first reported by Briene and Branemark.[1] In their study, 18 patients underwent the placement in the jaws of titanium implants surrounded by tibial cancellous bone chips. The authors chose not to use block grafts because of the belief that these grafts are really "dead" tissue with little likelihood of reestablishing vascularity. A healing period of 6 to 12 months was used, after which significant resorption of the graft occurred after loading; at 1 year after second stage surgery, only 25% of the implants remained integrated.

Listrom and Symington[2] placed implants simultaneously with several types of bone grafts to restore atrophied mandibles. The authors emphasized the need to place the implant into host bone as well as into the graft to maintain graft stability, and the need to use a

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healing period of 6 to 9 months. Additionally, they suggested that particulate bone grafts were difficult to pack around the implants and could contribute to their instability.

More recently, a surgical approach combining use of corticocancellous bone blocks in conjunction with implants has been used for restoration of the severely atrophied maxilla. In 1987, Keller et al[3] reported their findings with immediate and delayed placement of implants into corticocancellous bone blocks grafted in the maxilla. In the five patients receiving implants immediately after graft placement, 4 of 28 implants failed to integrate, whereas in the four patients who received implants after a 6 to 18-month graft healing period, 5 of 21 implants failed to integrate. The results of this study suggested that the bone grafts should be corticocancellous in nature; an adequate, but undefined, healing period for graft acceptance and implant integration should be used; and the implants should be placed in such a way that they engage both the graft and the host bone. Kahnberg et al[4] reported similar findings in a study of 57 implants in 10 patients in whom second stage treatment (abutment placement) was performed at 6 months after implantation. Eight of the 57 implants failed either because of initial instability of the implants or their exposure to the oral cavity.

Lew et al[5] used a 6-month healing period after simultaneous placement of corticocancellous block grafts and 43 implants in the mandibles of 10 patients. Using an external surgical approach, a 93% success rate was achieved with negligible bone loss at a 3-year follow up period.

In 1990, Adell et al[6] conducted a study using immediate autogenous corticocancellous grafts with implants in 23 patients (124 implants). A mean healing period of 9 months was used before loading. At 5 years, 74% of the original implants remained stable. Implant loss was attributed to failed osseointegration, not gradual grafted bone loss, and was probably related to a lack of rapid revascularization and remodeling of the graft. In a more recent study, Isaksson and Alberius[7] reported on eight patients undergoing simultaneous reconstruction of the atrophic maxilla and implant placement. They waited for 6 to 9 months before initiating the second stage of the implant procedure. At an evaluation period ranging from 32 to 64 months, 83% of the implants were well integrated.

Based on these previous clinical studies, there appears to be no unanimity as to the length of the healing period for the graft and implant before the second stage placement of the abutment. It is also apparent that the rate of osseointegration of titanium implants when placed in autogenous bone grafts has not been adequately investigated on an experimental basis. The objective of this work, therefore, was to compare the rate and extent of osseointegration of implants when placed simultaneously with either corticocancellous block or particulate corticocancellous chips using the canine iliac crest as the site for investigation.

Materials and Methods

Surgical and Implantation Procedure

Seventeen mongrel dogs (20 to 40 kg) were used in this investigation. After induction of general anesthesia delivered by an oral endotracheal tube, the dogs were secured to the operating table in a prone position with the femurs extended caudally. The surgical sites were shaved before induction and prepared for a percutaneous sterile surgical procedure. A 2% lidocaine solution with 1:100,000 epinephrine was used to infiltrate the surgical site. A 6-cm incision was made over the iliac crest using a #15 Bard Parker blade. Electrocautery in the cutting mode was used to incise the subcutaneous tissue, gluteal fat, and the superficial thoracolumbar fascia. The intermuscular septum between the superficial gluteal and middle gluteal muscles was then incised. Using a #15 Bard Parker blade, an incision was made in the crestal origin of the middle gluteal muscle on the tuber sacrale, starting at the cranial dorsal iliac spine and extending caudally for approximately 6 cm. An elevator was used to strip the periosteum and the overlying middle and deep gluteal muscles laterally and the iliocostalis and longissimus lumborum muscles medially. The crest and the wing of the ilium were then exposed inferiorly for a distance of approximately 2 cm. Using a #701 fissure bur mounted in a power drill, a vertical cortical cut was made 1 cm caudal to the cranial dorsal iliac spine. The cut was completed using an oscillating saw, proceeding inferiorly for a distance of 1 cm. It was then carried cranially for approximately 1 cm, completing sectioning of the spine. The specimen was placed in a sterile, saline-soaked gauze.

The procedure was repeated on the opposite side and a 1-cm corticocancellous block was resected, which was cut into 2 to 3-mm pieces using fine rongeurs. If the amount necessary for the graft was insufficient, more bone was harvested by resecting the crest in a ventrocephalic direction.

A wire-passing bur was next used to make a bicortical hole in the bone inferior to the graft receptor site on the second side and a 26-gauge wire was passed through the hole. A fissure bur was used to groove the block graft from the opposite side in a mediolateral direction on its superior border. The corticocancellous block was placed into the contralateral hip defect and the wire was tightened into the surface groove, thus immobilizing the graft against the host bone (Fig 1). Another hole was then made and a wire passed in a similar fashion around the intact host bone approximately 1 mm from

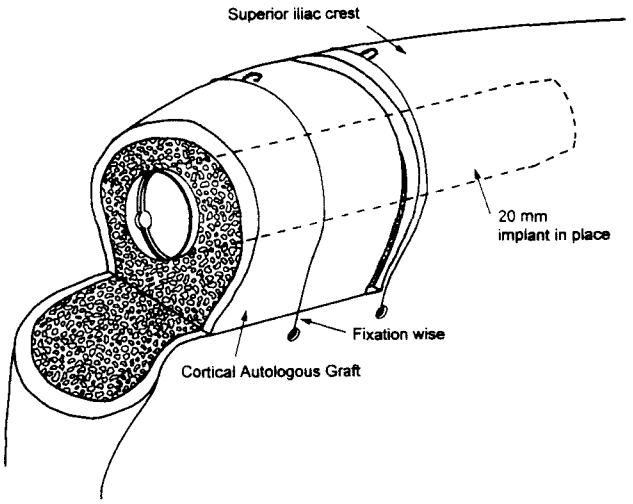

FIGURE 1. Schematic diagram of canine iliac crest used as the model in this study.

the junction of the graft and the unoperated receptor site bone. This wire was placed as a radiographic marker.

A self-tapping Branemark implant (20 mm length, 3.75 mm diameter) was placed horizontally through the corticocancellous block graft and into the adjacent host bone, and a cover screw was added. Approximately half the length of the implant was placed in the graft and the other half in the host bone as shown in Figure 1. The implant site was then closed in a layered fashion with Vicryl sutures for the deep layers and skin. Because 10 mm of every 20 mm implant was embedded in host bone, this portion of the implant served as an internal control for histologic analyses. In every case, this technique provided initial stability of the implant after placement. All implants were stable at the time of retrieval and were osseointegrated to varying degrees in the experimental and control portion of each implant site as determined by histologic analysis.

On the contralateral side, a Branemark implant was placed horizontally into the host bone for approximately half of its length in the same manner as on the opposite side. The non-implanted portion of the implant occupied a position ½ cm above the base of the previously removed iliac crest section. Thus, sufficient room was available to completely surround the implant with corticocancellous bone chips. These were placed firmly around the implant and the periosteum was sutured to cover the graft site using continuous 3–0 Vicryl sutures. Using this technique, a triangular-shaped pocket was created, the floor being the host bone and the lateral walls consisting of the periosteum sutured superiorly. Cancellous and cortical bone chips were packed into the pocket until the implant was completely surrounded by the grafted bone. Closure of the host site was performed in layers using continuous 3–0 Vicryl sutures.

Butorphanol (0.2 to 0.5 mg/kg, subcutaneously) was administered postoperatively to relieve pain. Antibiotic therapy (Ditrim [Syntex, Palo Alto, CA]; trimethoprim/sulfadiazine, 1 mL/20 lb, subcutaneously) was initiated 1 hour before surgery and then twice daily for 3 to 5 days. This was followed with the oral form, 120/160 mg daily for a total course of 10 days. The implants were retrieved for six dogs at 1 and 2 months and for five dogs at 3 months.

Histological Procedure

Oral tetracycline (100 mg/kg) was administered to three animals in each period to evaluate bone remodeling in the grafts and host bone adjacent to the implant. In every case, the tetracycline was administered within 48 hours of the surgery and 1 week before killing. At the appropriate times, the dogs were killed using an overdose of pentobarbital. The implant sites with surrounding bone were harvested by en bloc resection and the specimens were fixed in 10% neutral buffered formalin for 48 hours under vacuum. The tetracycline-labeled specimens were fixed in 70% ethanol at 4°C for 72 hours. All specimens were washed in three changes of phosphate-buffered saline for 1 hour. The specimens were then dehydrated in 70% ethanol followed by sequential immersion in 50%, 70%, 80%, and 95% glycol methacrylate (GMA) for 24 hours in each solution. Specimens were then placed in 2 changes of 100% GMA for 24 hours each under agitation. A 1:1 mixture of GMA and Technovit 7200 (T7200, Exakt Medical Instruments, Oklahoma City, OK) was used for 48 hours, with agitation to begin the infiltration process. Three changes of T7200, 72 hours for each change, were used to complete the infiltration. The specimens were embedded in the last change of T7200 with a gradual increase in temperature (to 40°C) to aid polymerization. Further polymerization was accomplished in a 60°C oven overnight.

Embedded specimens were fixed on Plexiglas slides and 150-μm thick sections were cut using a diamond saw blade on the Exakt cutting system. The sections were ground and polished to approximately 50 μm using the Exact grinding machine with water as the coolant.

For staining, the slides were agitated in 30% H_2O_2 for 5 minutes and then rinsed in tap water before staining with 1% Toluidine blue for 15 minutes. After staining, the slides were quickly rinsed in tap water and blotted dry. If further differentiation was required, the slides were briefly dipped in a 1:1 mixture of acetone-alcohol. The slides were air dried and temporarily coverslipped for viewing using immersion oil as the mounting media. The fluorescent markers were viewed using a Zeiss transmitted light photomicroscope with

an epifluorescence condenser, and a BP 450 excitor and LP 520 filter set with a xenon lamp.

Microradiographs of each wafer slide were prepared using Kodak 4489 EM film and a Faxitron x-ray cabinet (Field Emission Corp, McMinville, OR).[8] Photomicrographs were prepared of each wafer slide. The percentage of bone contact with the implant was determined by projecting the photomicroradiographs onto a digitizing pad and tracing the pertinent areas of bone contact. The percentages of bone/implant contact were determined for each of the most central wafer slides per implant site. Because each implant was placed into both graft and host bone, comparisons of the level of osseointegration as a function of graft material and host bone could be made on the bases of these wafer slides.[2] This procedure was performed by determining the midpoint (wire marker) of the implant in the wafer slide and measuring the bone-implant contact points both above (experimental) and below (control) the midpoint. The means and standard deviations of each experimental and control condition were calculated. Analysis of variance (two-way) with Duncan's Multiple Range tests ($P = .05$) were used to determine significant differences between the means.

Results

Histomorphometry

The data for the percent of bone/implant contact for each of the experimental and control conditions are presented in Table 1. It is apparent from this information that the percentage of bone contact (osseointegration) increased in the control portions of the implant sites as a function of time. However, there was no statistically significant difference ($P > .05$) between the particulate or block graft control group at each of the periods investigated. In the experimental particulate graft implant sites, there was a slight, but statistically insignificant ($P > .05$), increase in the percentage of osseointegration with time. This was attributed, in part, to the high degree of variability in osseointegration observed with this experimental group (coefficient of variation, 22% to 52%). The differences in percentage of osseointegration in the experimental particulate graft implant sites and the corresponding control sites were significant at both the 1 and 3 month periods ($P < .05$).

Table 1. Percent Bone Contact (Osseointegration) for Control and Grafted Implanted Sites

	Particulate Grafts		Block Grafts	
	Control	Experimental	Control	Experimental
1 Month (n = 6)	48.8 ± 15.0	26.1 ± 13.7*	54.8 ± 6.2	45.3 ± 7.0**
2 Months (n = 6)	59.8 ± 8.5	38.8 ± 12.6	56.0 ± 17.2	57.0 ± 7.6**
3 Months (n = 5)	63.2 ± 9.8	(39.2 ± 8.7)*	65.2 ± 8.3	(59.6 ± 4.2)

Single and double asterisks indicate significant differences in levels of osseointegration ($P < .05$) between experimental particulate graft sites (1, 3 months) and experimental block graft sites (1, 2 months), respectively. Parenthesis indicate significant differences between experimental particulate and block graft sites at 3 months.

In the block graft implant sites, the percentage of osseointegration also increased over time in both the graft implant sites and in the control sites. The differences between the control values at each period were not significant ($P > .05$); however, in the block graft implant sites, a significant ($P < .05$) difference in osseointegration was noted between 1 and 2 months. The difference in the levels of bone contact between the control and experimental block graft implant groups was also not statistically significant ($P > .05$) at any of the periods. The percentage of osseointegration for block graft implant sites was greater than for the particulate graft sites at each of the three periods investigated, and by 3 months this difference was statistically significant ($P < .05$).

Histological and Microradiographic Evaluation

The qualitative histologic examination of the prepared wafer slides provided additional clues as to the performance of the individual grafts in promoting osseointegration. It was apparent that both the particulate and block grafts remained viable and became incorporated in, and remodeled with, the host bone. This observation was confirmed by the quantitative histomorphometric analyses of osseointegration (Table 1) and by tetracycline-labeled sections. The tetracycline fluorescent label (yellow) immediately adjacent to the threads of the implant is indicative of active bone remodeling in both the particulate (Fig 2A) and block (Fig 2B) graft implant sites. For each type of graft implant site, the control portion of the implant was encased in varying amounts of host bone, which increased over time (Fig 3A, B).

In the experimental portion of each graft site, the general pattern of bone remodeling and interaction at the titanium interface is shown in Figures 4 and 5. At 1 month, bone remodeling was apparent adjacent to both the particulate (Fig 4A) and block (Fig 4B) grafted implant sites. In general, however, the implants were osseointegrated to a greater extent both qualitatively (Fig 4B) and quantitatively (Table 1) in the block bone grafts. Although a greater level of osseointegration was obtained in the particulate graft sites at 3 months than at 1 month (Fig 5A), the level of osseointegration was substantially greater, both qualitatively (Fig 5B) and quantitatively (Table 1) in the block graft implant sites.

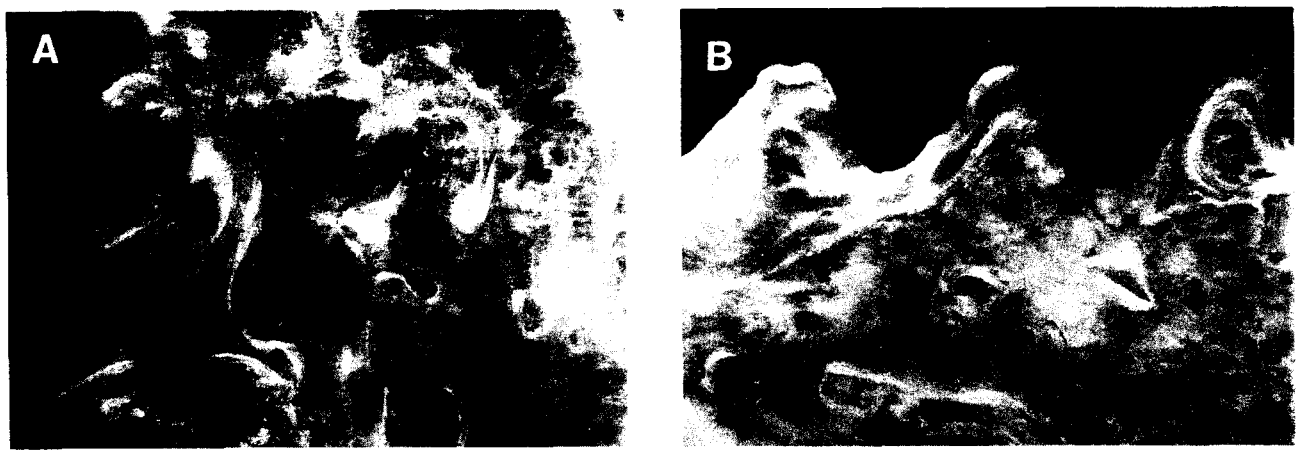

FIGURE 2. *A*, Photomicrograph of fluorescent (tetracycline)-labeled particulate graft/implant site at 3-month retrieval. *B*, Photomicrograph of fluorescent (tetracycline)-labeled block graft/implant site at 3-month retrieval. Yellow label adjacent to implant threads indicates active bone remodeling in both sections. (Original magnification ×10).

Discussion

The use of the canine iliac crest model proved effective for comparing bone graft responses at the implant sites. Using this model, each implant provided an experimental as well as a control component and the iliac crest site provided an adequate amount of graft material for both the cortical block and the particulate grafts.

The results of this study indicated that implants in the cortical block and the particulate grafts osseointegrate at different rates. By 1 month the cortical block implants demonstrated approximately 45% osseointegration, whereas the particulate graft implants demonstrated only 26% osseointegration (Table 1). This approximately 20% difference was mirrored in the 2- and 3-month results and was statistically significant at the 3-month period.

It has been suggested that approximately 40% osseointegration is sufficient to maintain a stable and clinically functioning implant.[9-12] Roberts'[13] studies indicated that the bone healing response in canines occurred at approximately *twice* the rate of humans. Therefore, this would suggest that from our results one could expect a clinically functioning implant in humans to be achievable at 2 months using cortical block grafts, whereas it would take 4 to 6 months to achieve this level of integration with particulate grafts. From the preceding results one can additionally infer that there was a faster rate of osseointegration with corticocancellous block graft than with one consisting of corticocancellous chips, the ratio being approximately 3 to 1.

The reasons for the results obtained would seem to revolve about three considerations: 1) the degree of vascularity of each graft, 2) the degree of trauma in

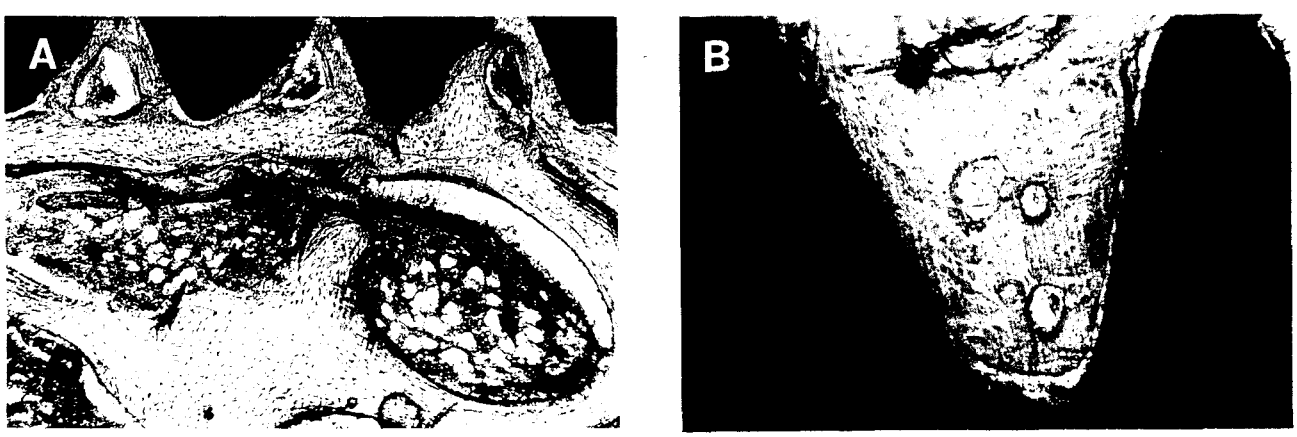

FIGURE 3. *A*, Control portion of particulate graft implant site at 1-month retrieval. Note ingrowth of bone into threaded portions of implant site (Toluidine blue stain, original magnification ×16) *B*, Higher magnification of control portion of block graft implant site at 1-month retrieval demonstrating significant ingrowth of bone into the threaded portion of the implant and areas of apparent contact of bone with the implant surface. (Toluidine blue stain, original magnification ×40).

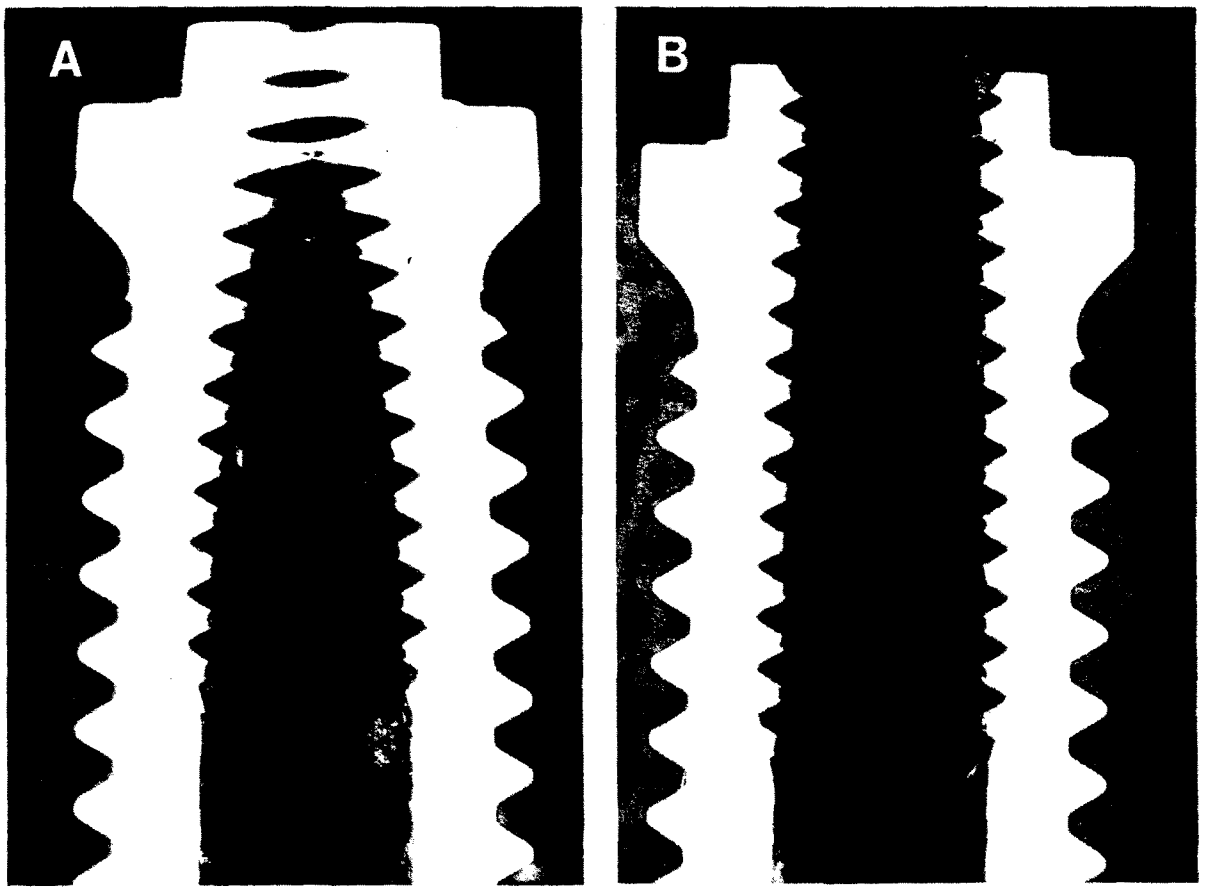

FIGURE 4. *A*, Microradiograph of experimental portion of a particulate graft implant site at 1-month retrieval. Note bone remodeling of the graft adjacent to the implant. Sparse bone contact with the threads. *B*, Microradiograph of experimental portion of a block graft implant site at 1-month retrieval. Note more significant bone remodeling of the graft and contact with the implant threads. (Original magnification ×5).

preparation of the graft, and 3) the initial degree of bone-implant interface.

Determining the degree of vascularity was not an objective for study in this experiment. The literature does note that cancellous bone grafts have a faster revascularization rate when compared with corticocancellous grafts.[1,14,15] The revascularization phenomenon is, indeed, an early one in the bone healing cascade, occurring before the initial sacrifice period used in this work (1 month).[15] It should be pointed out that the grafts were positioned so that vascularization was obtained from two surrounding host medullary bone walls. This factor may have contributed to the accelerated bone response observed with the block grafts. We recognize, however, that in some clinical situations there is only a single host bone graft interface as would occur with onlay graft.

Albrektsson[14] has noted that "minimization of trauma to autogenous bone grafts results in more rapid revascularization and bone remodeling of the graft at the host site."[14] Considering this hypothesis, it would appear that preparation of the particulate corticocancellous grafts was more traumatic than harvesting of the cortical block. One assumes that this would be related to the relative number of viable osteocytes remaining in the graft and graft site. The work of Sennerby et al[16] has indicated a higher degree of implant retention when there was initially a greater degree of contact between the host bone and the implant surface. Clearly, this would be the case when the implant is placed into a corticocancellous block graft rather than into particulate graft material. It is of further interest that the degree of osseointegration was not statistically different in the cortical block graft site and in the control site throughout the 3-month period of investigation.

This experiment has demonstrated a higher rate and degree of osseointegration when an autogenous corticocancellous block grafts is used in conjunction with implants than when a particulate corticocancellous graft is used. These results suggest that, when faced with a choice between the use of the two grafts, the block graft may result in a hastening of the osseointegration process.

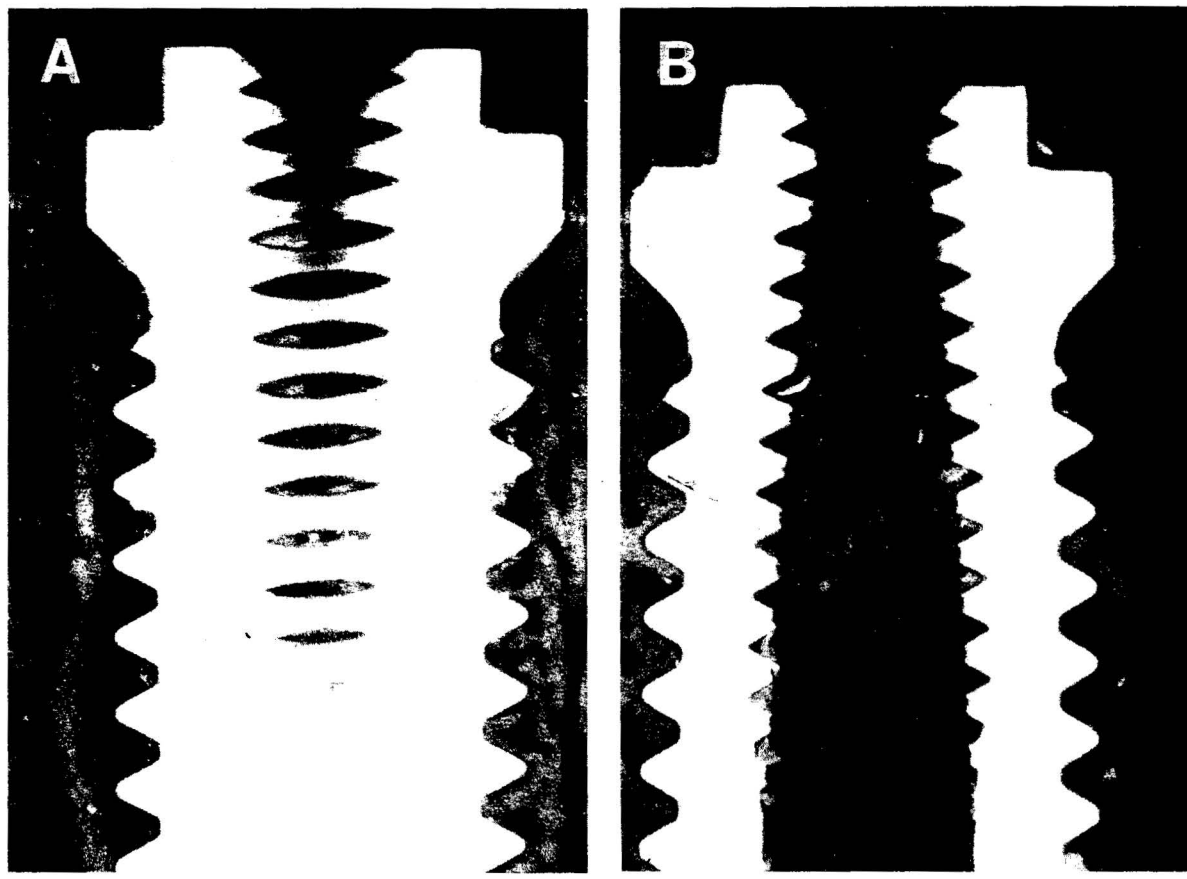

FIGURE 5. *A*, Microradiograph of experimental portion of particulate graft implant site at 3-month retrieval. Bone remodeling of graft is seen adjacent to and within the threaded region of the implant (original magnification ×5). *B*, Microradiograph of experimental portion of block graft implant site at 3-month retrieval. Note how bone in the graft site has contacted the threaded portion of the implant (original magnification ×5).

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J Oral Maxillofac Surg
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Discussion

A Comparative Study of Osseointegration of Titanium Implants in Corticocancellous Block and Corticocancellous Chip Grafts in Canine Ilium

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Lew et al have developed a logical, thoughtful review of the literature and, based on previous reports and unresolved issues, have designed their study with the objective of comparing the rate and extent of osseointegration of implants using two physically different compositions of autografts: block and particulate. The dog wound model used is clever and appears suitable. Using this model, 20 mm × 3.75 mm-diameter self-tapping Branemark implants were placed either through the corticocancellous block that was fixed with wires to the host or the implants were placed directly into the donor bed and then augmented with particulate bone autograft. The authors stated that this relationship between implant and bone resulted in each implant serving as its own control (the 10 mm of screw length embedded into the host bone). Ideally, the experimental and control implants should have equivalent lengths and locations, and the same locale on the implants should be assessed. However, there seems little way around this detail, and one may only speculate whether minor differences in function could occur along different lengths of the implants. The implant site is "passive"; consequently, this point may be irrelevant.

Tetracycline was administered to the dogs. Histologically, the authors equated vital bone with fluorescence of the tetracyclines bound to the bone. This is an acceptable marker for bone mineralization and, together with morphology, it was used appropriately to identify zones of remodeling.

The authors measured bone-implant contact using quantitative imaging of radiographs made from thin sections through implant and bone. Using this technique, they determined a greater percent bone-implant contact for blocks than particulate grafts. In addition, tetracycline fluorescence of the bone-implant front confirmed viable bone. Based on percent block-implant data, the authors concluded the bone formation rate was more rapid in the block than in the particulate grafts. This conclusion may be a bit of a stretch simply because bone formation rate is defined as a unit measurement per unit time.[1,2] Classically, distance between fluorescing tetracycline bands is measured, and knowing the temporal difference between administered doses of tetracycline, the mineral apposition rate is derived. This exercise was not accomplished by the authors. However, a valid conclusion can be made that there was more bone along the implant within the block than the implant surrounded by the particulate graft. Did it form more quickly or was there more initial contact at the start? Table 1 appears to support this notion.

The authors posited results were based on three variables: vascularity, trauma, and initial bone-implant interface. These appear valid. I would speculate that the physical property of the two graft preparations, particulate versus block, was probably the most important determinant for osseointegration. Particulate graft placed next to the implant will undergo a predictable sequence of responses modulated by cells and local regulatory molecules. These will include resorption, activation, formation, and consolidation.[3,4] The sensors and effectors modulating these local responses will be influenced by function.[5] Therefore, one may question the virtue of a passive wound model. With what degree of fidelity does it relate to the alveolar bone? This question is neither posed to detract from the merit of the study nor to discount the results and conclusions. It is posed as an invitation to design an experimental wound model that may be more functionally relevant to alveolar bone.

Based on comments vide supra, a question to pose is whether the investigators gave the implant in the cortical block a "head-start" on osseointegration? The block recipient bed already had a degree of physical-structural organization allowing for immediate bone-implant contact. Does this invalidate the authors' conclusion? I do not believe it does. It is supportive. Moreover, a head-start will be a valuable clinical asset for patients with systemic liabilities such as postmenopausal osteoporosis.

A final comment on the animal wound model focuses on the embryologic derivation of the autograft and recipient bed. Both are endochondral. Can we predict a similar response at an intramembraneously-derived host bed (eg, the alveolar bone)? This is another invitation to design an embrylogically relevant study to verify the observations reported by Lew et al.

In conclusion, the work by Lew et al is a valuable contribution to the literature. Additional, carefully designed, well-executed studies such as this one need to be accomplished to validate observations. I congratulate the authors on their efforts.

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