Radiographic Evaluation of Bone Adaptation Adjacent to Percutaneous Osseointegrated Prostheses in a Sheep Model

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Abstract

Background Percutaneous osseointegrated prostheses (POPs) are being investigated as an alternative to conventional socket suspension and require a radiographic followup in translational studies to confirm that design objectives are being met.

Questions/purposes In this 12-month animal study, we determined (1) radiographic signs of osseointegration and (2) radiographic signs of periprosthetic bone hypertrophy and resorption (adaptation) and (3) confirmed them with the histologic evidence of host bone osseointegration and adaptation around a novel, distally porous-coated titanium POP with a collar.

Methods A POP device was designed to fit the right metacarpal bone of sheep. Amputation and implantation surgeries (n = 14) were performed, and plane-film

radiographs were collected quarterly for 12 months. Radiographs were assessed for osseointegration (fixation) and bone adaptation (resorption and hypertrophy). The cortical wall and medullary canal widths were used to compute the cortical index and expressed as a percentage. Based on the cortical index changes and histologic evaluations, bone adaptation was quantified.

Results Radiographic data showed signs of osseointegration including those with incomplete seating against the collar attachment. Cortical index data indicated distal cortical wall thinning if the collar was not seated distally. When implants were bound proximally, bone resorbed distally and the diaphyseal cortex hypertrophied.

Conclusions Histopathologic evidence and cortical index measurements confirmed the radiographic indications of adaptation and osseointegration. Distal bone loading,

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through collar attachment and porous coating, limited the distal bone resorption.

Clinical Relevance Serial radiographic studies, in either animal models or preclinical trials for new POP devices, will help to determine which designs are likely to be safe over time and avoid implant failures.

Introduction

Percutaneous osseointegrated prosthetic (POP) devices connect an artificial limb directly to the residual stump bone via an osseointegrated intramedullary implant. The many advantages of skeletal docking have supported advancement of this system as an important potential alternative to conventional socket suspension. Particularly suited to patients with short stumps and multiple limb loss, POP devices are designed to transmit ground reaction forces directly to the skeletal system. They provide the advantages of osseoperception and improved gait symmetry while eliminating socket-induced stump pain, skin pathologies, and fitting problems inherent to socket suspension [12, 13, 21]. For more than 10 years, three different POP designs have been implanted in European amputee volunteers: the osseointegrated prosthesis for the rehabilitation of amputees (OPRAs) in Sweden (Fig. 1A) [11], the endo-exo femur prosthesis in Germany (Fig. 1B), and the intraosseous transcuteaneous amputation prosthesis (ITAP) in England (Fig. 1C) [17] [2].

Each implant system has unique endoprosthetic surface designs to achieve osseointegration. While the OPRA design uses a threaded titanium alloy cylinder that is screwed into the reamed medullary canal [11], the endo-exo femur prosthesis is a cobalt-chrome shaft curved to accommodate the anterior femoral bow (which provides rotational stability) with a cast-structured tripod outer layer for bone ingrowth [2]. To supplement rotational stability, the intramedullary component of the ITAP is a fluted titanium alloy cylinder [17] (Fig. 1C). All three designs attempt to achieve the transfer of ground reaction forces directly to the residual bone through an optimally osseointegrated intramedullary implant. They also attempt to conserve the host bone tissue and minimize stress shielding to avoid any adverse bone adaptation. Although good medium-term survival rates have been reported for these European implants [2, 11], to our knowledge, there has only been one publication that used sequential radiographic evaluation of patients to attempt to ascertain the effect of skeletal docking on bone remodeling over extended time [19]. Without serial clinical radiographic followup, it is impossible to determine whether the design

objectives of limiting physiologic bone resorption and implant loosening are being met over the life of the implant [19]. This knowledge is essential if late catastrophic implant failure, due to initially faulty implant designs, is to be avoided.

Periprosthetic bone remodeling and adaptation can be attributed to the geometry, surface design features, implant material, and surgical placement of the device. In patients implanted with the OPRA system [19], although radiographic data indicated stable fixation of the POP devices, proximal “cancellization” of cortical bone and pronounced distal bone resorption were seen in more than 1/2 of the patients at 2-year followup. Based on bone remodeling principles [7, 22, 23], it can be inferred that the screw-in design and placement of the OPRA implant inherently failed to load the distal region of the bone (2 cm of distal bone, Fig. 1A) and likely accounts for the observed extensive distal bone resorption. It is therefore inferred that implant constructs with bone end loading collars and distally porous-coated shafts for assuring distal bone loading would minimize distal bone resorption and optimize osseointegration. This forms the rationale for the implant design reported on in this study (Fig. 2). This novel implant design was engineered to optimally fit and fill the anatomic dimensions of the distally transected sheep metacarpal and was tapered to avoid loading at the proximal medullary canal. The intention was to ensure distal load transfer and the hoped for evenly acquired hypertrophy along the proximal length of the bone.

In this 12-month animal study, we determined (1) radiographic signs of osseointegration and (2) radiographic signs of periprosthetic bone hypertrophy and resorption (adaptation) and (3) confirmed them with the histologic evidence of host bone osseointegration and adaptation around a novel, distally porous-coated titanium POP with a collar.

Materials and Methods

Animal Surgery

Using institutionally approved Institutional Animal Care and Use Committee protocols from the US Department of Defense, Salt Lake City Veterans Affairs (Salt Lake City, UT, USA), and Innovative Medical Device Solution Discovery Research (Logan, UT, USA), a single-stage amputation and implantation procedure was performed on 14 2- to 4-year-old, skeletally mature, mixed-breed sheep (Table 1). Each sheep was implanted with a novel porous-coated (P²; porosity = 52% ± 12%; Thortex Inc, Portland, OR, USA) endoprosthesis design (Fig. 2). After implantation surgery, the operative site was protected with a

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Fig. 1A–C Schematic representations show POP devices (not drawn to scale) clinically used in European countries. (A) Swedish design (OPRA); (B) German design (endo-exo femur prosthesis); and (C)

British design (ITAP). 1 = endoprosthesis; 2 = host bone tissues; 3 = soft tissues; 4 = exoprosthesis; 5 = subdermal barrier for improving soft tissue integration.

Fig. 2A–B A schematic representation shows host bone tissue surrounding the POP used in this study: (A) AP and (B) ML views. Each image was delineated into seven (14 in total) zones for the evaluation of bone resorption and the presence of radiolucent lines. The length of each zone was specific to the topography features of the implant.

Table 1. Demographic data of study animals and implant sizes used to achieve a press-fit within the intramedullary canal

Animal	Age (years)	Breed	Sex	Implant size (number)	Cerclage wire used?
1	3	Rambouillet	Female	2	No
2	4	Rambouillet	Female	1	No
3	2–3	Targhee	Female	2	No
4	2	Polypay	Female	3	No
5	2	Crossbred	Female	2	No
6	2–3	Suffolk	Female	1	No
7	2–3	Targhee	Female	1	Yes
8	2–3	Targhee	Female	1	No
9	2–3	Targhee	Female	1	Yes
10	3	Polypay	Female	1	No
11	2–3	Suffolk	Male	2	No
12	2–3	Suffolk	Female	1	No
13	2–3	Suffolk	Male	3	No
14	2–3	Suffolk	Male	3	No

surgical dressing for 2 weeks, and the animals were allowed to freely ambulate for 12 months [20]. A full description of the surgical procedure was published elsewhere [15, 20].

Postoperative Animal Care

For 3 weeks postimplantation, all animals were housed separately within internal pens and then moved to a

communal outside pen for the remainder of the study. To minimize the bioburden at the interface, skin tissues surrounding the percutaneous post were cleaned weekly with an antimicrobial spray (benzethonium chloride; Pure-Works[®]; Pure Works, LLC, Farmington, UT, USA) and observed for superficial and systemic signs of infection. The implant site was also observed for clinical signs of bone fractures, lameness, and implant loosening. The sheep were euthanized at the end of the 12-month period and were subjected to histologic evaluation. Euthanasia was performed in accordance with 2007 American Veterinary Medical Association guidelines.

Radiographic Followup and Evaluation

A set of AP and mediolateral (ML) radiographs were taken postoperatively at 0, 3, 6, 9, and 12 months using a portable x-ray unit (PXP-40HF; United Radiology Systems Inc, Deerfield, IL, USA) to monitor the bone-to-implant integration (osseointegration) and for documenting bone adaptation. Three blinded observers (SJ, OC, LB) assessed these plane radiographs. The observers agreed with each other's assessments the majority of the time, but when observations were dissimilar, microradiographs were used for clarification and/or an experienced orthopaedic surgeon (JPB) was consulted.

Signs of Osseointegration

In line with the work of Engh et al. [6], the absence of a neocortex (reactive lines; Fig. 3, black arrows) and radiolucent lines adjacent to the porous-coated region and the presence of endosteal new bone or spot welds were considered signs of osseointegration. To allow comparison between animals and to evaluate each animal over time, the serial radiographs were delineated into 14 zones (Fig. 2) based on surface features of the endoprosthesis [5, 10, 19]. The porous-coated zones (Zones 1, 7, 8, and 14; Fig. 2) were examined for the presence or absence of radiolucent lines and neocortex, and the fluted and smooth regions (Zones 2, 3, 5, 6, 9, 10, 12, and 13; Fig. 2) were evaluated for the presence of spot welds.

All zones were also assessed for the intimacy of implant contact to the bone (apposition) or conversely radiolucent lines. When a radiolucent line was present or became apparent in any zone, its progression was assessed over the entire study period. For the semiquantitative data analyses, radiolucent lines involving 50% or more of the respective zones were considered nonosseointegrated zones [24]. The number of implant contact zones were counted and compared between Time 0 and 12 months.

Fig. 3 An annotated radiograph (AP view) shows the reference positions used for computing cortical index measurement: proximal end of the porous-coated region of the endoprosthesis (Line 1), proximal end of the fluted region of the endoprosthesis (Line 2), and proximal implant tip (Line 3). Based on the topography of the implant design, periprosthetic bone was divided into three specific regions: porous-coated region (A), fluted region (B), and smooth region (C). Black arrows point to the neocortex formation/reactive line, which is defined as the demarcation of radiolucent lines with a secondary bone ring.

Quantification of Bone Hypertrophy and Resorption (Cortical Index Measurements)

To quantify the host bone hypertrophy and resorption, the cortical index value was computed from the AP and ML

radiographs taken at Time 0 and 12 months. The cortical index was calculated using the following equation [9] and expressed as a percentage:

$$\text{cortical index} = \frac{\text{SW} - \text{MW}}{\text{SW}} \times 100$$

where SW is the metacarpal shaft width measured in millimeters and MW is the medullary canal width measured in millimeters. The diameter of the stem was used as a scale marker. The mean cortical index values were calculated from three consecutive measurements per zone by an orthopaedic fellow (OC).

For these measurements, the radiographic films were digitalized using a scanner (Umax Powerlook 2100XL; Umax Technologies Inc, Dallas, TX, USA) with Magic Scan software (Umax Technologies Inc). An IMAQ™ Vision Builder software (National Instruments, Austin, TX, USA) was then used to measure the width of the metacarpal shaft (SW) and the width of the medullary canal (MW) at three defined locations: (1) the proximal end of the porous-coated region of the endoprosthesis (Fig. 3, Line 1), (2) the proximal end of the fluted region of the endoprosthesis (Fig. 3, Line 2), and (3) the proximal implant tip of the smooth region of the endoprosthesis (Fig. 3, Line 3). The cortical index changes over this 12-month period were then calculated to determine the thickening or thinning of the cortical wall.

Assessment of Implant Collar Seating

The presence or absence of initial contact between implant and resected bone cross section was assessed from the

postimplantation (Time 0) radiographs. The bone tissue responses to the initial seating, including distal rounding off and distal bone resorption, were assessed from the 12-month radiographs. Although not always achieved, direct contact of the implant with the transected bone end was considered important for conserving the distal bone structure (Fig. 4, Zones 15–18) by assuring that axial loads transferred from the implant to the resected bone surface and endosteum at these designated porous-coated distal regions. This was an important design intention of this particular implant construct.

Histopathologic Evaluations

At necropsy, seven of the 14 specimens were harvested and subjected to mechanical testing and reported elsewhere [16]. Using an established method, the remaining seven specimens [3 sections per each region (porous, fluted and smooth regions; Fig. 2)] were processed for histologic analyses [14, 15].

Scanning Electron Micrographic Analysis

Representative sets of polished sections from each of the zones were evaluated for micrographic evidence of periprosthetic bone integration with the implant surface. They were further assessed for bone types (cancellous, cortical, or woven) using a scanning electron microscope (SEM) (JSM 6100; JEOL Inc, Peabody, MA, USA) with an attached high-resolution backscattered electron (BSE) detector (Tetra; Oxford Instruments, Cambridge, UK) at x20 and x100 magnification [3].

Fig. 4A–B Distal view of (A) AP and (B) ML radiographs of POP devices implanted into sheep metacarpal bones showing Zones 15 to 18, which were used for assessing the direct load transfer (seating) of the implant collar at the transected distal end.

Histologic Analyses

Representative sections were analyzed for evidence of osseointegration using histologic staining techniques. For this, at least one section from each area was ground to 50- to 75- μ m thickness and polished (EcoMet[®] 250 Grinder-Polisher; Buehler, Lake Bluff, IL, USA) to acquire an optical finish. The sections were then immersed in a preheated (50°–55° C) solution of Sanderson's Rapid Bone Stain[™] (Dorn and Hart Microedge, Villa Park, IL, USA) for 2 minutes, followed by rinsing in running water for 10 minutes. The specimens were then counterstained with acid fuchsin for 15 minutes at room temperature and imaged using a Nikon Eclipse E600 microscope (Nikon Corp, Tokyo, Japan) with an attached PAXcam[™] digital camera (MIS Inc, Villa Park, IL, USA). PAX-it[™] image analysis software (MIS Inc) was used for still image capture of the specimens.

Results

Radiographic Signs of Osseointegration

At Time 0, radiographs from all 14 animals showed intimate contact (apposition) of bone with the endoprosthesis in at least five zones (Table 2). By 12 months postimplantation (Table 2), five of the 14 animals had increased the number of apposition zones, while six had maintained their initial number of apposition zones. The remaining three animals had decreased the number of initial apposition at specific zones within the fluted and smooth regions. In some cases ($n = 5$), periprosthetic bone tissue was overreamed in the fluted and smooth regions, which resulted in the presence of radiolucent lines. These overreamed fluted region (Zones 2, 6, 9, and 13) appeared to fill in with new bone tissue or spot welds with increasing implant in situ time, which was a sign of regional osseointegration.

At 12 months, all radiographic data showed evidence of osseointegration (Table 3), where there was no radiolucent line or necortex present in the distal porous region (Zones 1, 7, 8, and 14). Moreover, all but one animal (Animal 7) showed radiographic signs of spot welds or endosteal new bone in the proximal fluted or smooth regions (Zones 2, 3, 5, 6, 9, 10, 12, and 13). In the single animal without proximal spot welds (Animal 7), an immediate tight fit and fill of the implant had been achieved, with minimal to no gap between the implant surface and bone tissue, indicating endosteal loading at Time 0. The 12-month radiographs of 10 animals (Table 3) showed the presence of radiolucent lines in the fluted and smooth regions, but they had not

Table 2. Number of implant-to-bone apposition/contact zones with increasing implant in situ time and changes to bone-to-implant apposition zones between Time 0 and 12 months postimplantation

Animal	Number of implant-to-bone apposition/contact zones					
	Postsurgery	3 months	6 months	9 months	12 months	Change
1	7	6	6	8	10	+3
2	6	6	7	8	8	+2
3	5	6	5	5	5	0
4	8	8	8	Not taken	8	0
5	9	10	10	Not taken	12	+3
6	6	6	6	4	3	−3
7	12	11	11	12	12	0
8	5	5	6	6	6	+1
9	5	5	5	5	5	0
10	8	8	7	7	8	0
11	6	6	6	6	6	0
12	9	9	7	7	7	−2
13	5	7	8	9	9	+4
14	9	9	7	7	7	−2
Mean $\pm$ SD	7 $\pm$ 2				8 $\pm$ 1	

progressed from Time 0 except in a single zone (Zone 2) of Animal 2.

Radiographic Signs of Periprosthetic Bone Resorption and Hypertrophy (Adaptation)

Changes in cortical index data were used to quantify the overall bone adaptation (Fig. 5). These data suggested that there was generalized cortical thickening of host bone tissue, especially in the diaphyseal cortex (Fig. 5, ML 2 and AP 2). However, in eight animals, distal ML cortical wall thinning was observed, though cortical wall thinning exceeded 5% in only three of these animals. These latter data were in line with the radiographic image showing prominent distal rounding off and bone resorption (Fig. 6). At the proximal end of the implant (Fig. 3, Line 3), four animals showed minimal (< 3%) bone resorption in the AP direction.

In four animals (Animals 5, 7, 12, and 14), because of mismatch between implant size and canal anatomy, no contact of the implant collar with the transected bone end was achieved (Table 4). This resulted in pronounced distal bone resorption by 12 months (Fig. 6). When the anterior, posterior, medial, and lateral regions of the transected bone

Table 3. Radiographic indications of osseointegration along the length of the endoprostheses at 12 months postimplantation

Animal	Presence of reactive lines/neocortex in porous-coated region (Zones 1, 7, 8, and 14)	Presence of spot welds in fluted and smooth regions (Zones 2, 3, 5, 6, 9, 10, 12, and 13)	Spot weld zones	Radiolucent lines	Radiolucent line progressive?
1	No	Yes	2, 6, 13	3, 5, 12	No
2	No	Yes	6, 13	2, 3, 5, 12, 13	No
3	No	Yes	3, 6, 13	2, 3, 5, 6, 12, 13	Yes (only in Zone 2)
4	No	Yes	6, 13	10	No
5	No	Yes	2, 6, 13		No
6	No	Yes	2, 6, 13		No
7	No	No			No
8	No	Yes	2, 6, 12, 13		No
9	No	Yes	6, 13	10	No
10	No	Yes	2, 3, 12, 13	3, 5, 10, 12	No
11	No	Yes	2, 3, 5, 6, 13	10, 12	No
12	No	Yes	2, 3, 5, 6, 12	13	No
13	No	Yes	2, 5, 6, 9, 10, 12, 13	3, 5	No
14	No	Yes	3, 5, 10, 13	5	No

Fig. 5 A bar chart shows the difference in the cortical index (CI) between the postsurgical and 12-month time points. Each cluster of six bars represents the cortical index changes between 0 and 12 months of a single animal calculated from AP and ML radiographs. Negative

values represent thinning of the cortical wall. Positive values represent thickening of the cortical wall. Measurements were made in triplicate: at the proximal implant tip (Line 3), at the proximal flute (Line 2), and at the proximal end of the porous-coated region (Line 1) (Fig. 3).

end were in contact with the implant collar (Animal 13), no bone resorption was noted (Fig. 7). Overall, six animals showed marked resorption in porous-coated zones (Table 4) and six animals showed distal ML rounding off. Two animals (Animals 7 and 9) had intraoperative cortical

bone fractures during implantation and required cerclage wires for stabilization. Whether or not these two implants were well seated, the use of these wires resulted in marked distal bone resorption (Fig. 8). Thus, implants with cerclage wires were excluded from this evaluation.

Fig. 6A–D A serial set of radiographs taken at (A) Time 0 and at (B) 3, (C) 6, and (D) 12 months postsurgery and BSE micrographs (12 months postsurgery) show a cross section from a representative case of a nonideal implant placement. Six- and 12-month radiographs clearly show a pronounced region of distal bone resorption (arrows).

The BSE images show a high degree of resorption at the distal end, where cancellous bone has been resorbed and remodeled into a cortical bone-type structure (Point 1). Porous coating and implant are shown in white, bone in gray, and cellular components in black.

Table 4. Radiographic evidence of postsurgical collar seating at Time 0 and associated distal bone responses at 12 months postimplantation

Animal	Time 0					12 months
	Number of collar seating zones	Anterior side seating (Zone 15)?	Posterior side seating (Zone 16)?	Medial side seating (Zone 17)?	Lateral side seating (Zone 18)?	
1	3	Yes	Yes	Yes	No	Rounding off
2	1	No	Yes	No	No	Rounding off
3	3	Yes	No	Yes	Yes	Rounding off
4	2	Yes	No	Yes	No	Resorption
5	0	No	No	No	No	Resorption
6	3	Yes	Yes	Yes	No	Resorption
7	0	No	No	No	No	Resorption
8	Not taken/bone fractured and cerclage wire was used					Resorption
9	3	Yes	Yes	No	Yes	Rounding off
10	Not taken/bone fractured and cerclage wire was used					Resorption
11	2	No	Yes	Yes	No	Rounding off
12	0	No	No	No	No	Resorption
13	4	Yes	Yes	Yes	Yes	Rounding off
14	0	No	No	No	No	Resorption

Histologic Signs of Periprosthetic Bone Osseointegration and Adaptation

All of the implants were integrated well with the host bone at the porous-coated region (Fig. 9A, D) and the implant

was stabilized by bony ingrowth in the fluted region (Fig. 9B, E). When distal resorption was present at 12 months, the metaphyseal cancellous bone present at Zones 1, 7, 8, and 14 (porous-coated region) had remodeled into cortical bone. These radiographic changes were confirmed

Fig. 7A–D A serial set of radiographs taken at (A) Time 0 and at (B) 3, (C) 6, and (D) 12 months and BSE micrographs show a cross section from a representative case of an ideally placed implant. The BSE images clearly show the presence of cancellous bone tissue at the

distal end, which indicates that the original bone structure has been preserved (Point 1). Porous coating and implant are shown in white, bone in gray, and cellular components in black.

Fig. 8A–C Representative 12-month (A) AP and (B) ML radiographs show a pronounced zone of resorption at the lateral side, where a cerclage wire was used. However, the implant is well secured with bone ingrowth at the AP regions, (C) which was confirmed using SEM.

by histopathologic evaluation (Fig. 6, Point 1; Fig. 9D). In addition, compared to properly fitted specimens (Fig. 9C), fibrous capsules were seen in the smooth region (Zones 3, 5, 10, and 12) of some specimens (Fig. 9F). Animals with a tight proximal implant fit became stress shielded distally and showed cortical bone hypertrophy at the point where the implant was bound in the proximal medullary canal. Moreover, histologic evidence confirmed new bone formation within the overreamed fluted region (Fig. 10).

Discussion

Periprosthetic bone adaptation is an inevitable response to osseointegrated endoprostheses. The long-term radiographic documentation of this adaptation, as well as the early evidence of osseointegration, is of particular clinical importance because they give an indication of the stability of the implant-to-bone construct. This documentation is also essential when validating the design intentions of new

Fig. 9A–F A representative set of stained bone-implant cross sections taken at (A, D) porous-coated, (B, E) fluted, and (C, F) smooth regions is shown for (A–C) an ideal case and (D–F) a tightly fitted case, where the implant failed to be seated at the resected end (oversized implant) (stain, Sanderson's Rapid Bone StainTM;

counterstain, acid fuchsin). (B, E) Good bone ingrowth is seen at the fluted region in both cases; however, when nonideal placement occurs, (D) cancellous bone is remodeled into cortical-type bone at the distal end and (F) fibrous encapsulation (blue) occurs in the proximal end. P = porous coating; I = implant.

Fig. 10 A representative stained cross section taken from an overreamed fluted area shows spot welds (bone bridging/ongrowth) without any fibrous capsule formation (stain, Sanderson's Rapid Bone StainTM; counterstain, acid fuchsin).

implant systems in translational animal models and pre-clinical trials. In this 12-month animal study, we determined (1) radiographic signs of osseointegration and (2) radiographic signs of periprosthetic bone hypertrophy and resorption (adaptation) and (3) confirmed them with the histologic evidence of host bone osseointegration and

adaptation around a novel, distally porous-coated titanium POP with a collar.

This study had limitations. First, it was difficult to target a focused rehabilitation program for these animals. However, in clinical settings, compliance and feedback from patients would be anticipated. This would allow one to control the degree of initial loading and to collect patient-reported outcomes, including patients' pain and tolerance levels to weightbearing. It is expected that, together with the clinically relevant patient-reported outcome measures, periodic radiographic followup data would validate and confirm the design principles of this implant system further. Secondly, a lack of adequate range of implant sizes prevented an initial press-fit in some cases, where implants were undersized and mechanically compromised. In other cases, oversized implants resulted either in fractures ($n = 2$) or in stress shielding ($n = 6$). In human trials, multiple implant sizes and/or custom designs would be advocated for ensuring ideal fitting. Thirdly, host bone factors also influence bone remodeling around the endoprostheses, which include subjects' general health, disuse bone osteopenia/osteoporosis, cortical wall thickness, and the geometry of the implant and the host bone. In this sheep model, the effect of host variability was limited by using animals of similar age. However, these factors could produce variable bone responses in human amputees with

different degrees of disuse bone osteopenia (bone density) and residual bone geometry compared to the active, 2- to 3-year-old sheep with uniform mechanical properties of bone. Finally, the followup period of this translational study was short, and longer-term followup is necessary to determine whether the observed bone responses would be exacerbated with increasing in situ time.

The data suggest that the features of the distal collar, anatomic shape, and distal porous coating helped to improve the osseointegration of this novel POP device. The data further suggest that when the implant was properly seated, distal resorption was minimized (Table 4). However, the limited number of implant sizes prevented some implants from being properly fitted and seated on the collar, leading to distal bone resorption and proximal hypertrophy caused by the proximal implant binding. This was clearly observed in the SEM images (Figs. 6, 7).

To achieve implant fixation within the canal of the right metacarpal of the sheep, the endoprosthetic component was anatomically tapered. As this implant is designed to achieve distal bone fixation, the distal 1-cm portion of the endoprosthetic component was porous coated. It was found that anatomic shape of the implant and the surgical protocols helped to achieve and maintain at least five contact/apposition zones (Table 3) throughout the length of the each implant. It was believed that these contact zones, mainly at the distal regions, might provide the initial stability to prevent micromotion-related implant loosening and help to achieve the robust osseointegration, which was demonstrated by pullout strength data in a related study [16]. The absence of radiolucent and neocortex formation within this porous region, observed in 12-month radiographs, indicated that the implant was osseointegrated within this region. Neocortex and radiolucent lines were, however, seen in the proximal portion (mainly smooth region) of the endoprostheses and did not necessarily equate to the failure of bone integration with the endoprosthesis, since these lines were not progressive in sequential radiographs (Table 4). The histologic signs of spot welds (Fig. 10) present within the fluted region of these specimens further confirmed implant fixation by regional osseointegration. Although the implant was intentionally porous coated to a length of only 1 cm of structure at the distal end, the pullout strength study of this device showed extremely robust osseointegration with approximately 1000-N pullout at Time 0 and exceeding 13,000 N at 12 months [16]. If this attachment strength can be achieved in the human situation, this particular POP design would likely be bone sparing in the event of revision surgeries that require implant removal. This is in comparison to another implant design in which the porous coating is used along the entire length of the implant [2], which can result in excessive bone loss when revised.

Bone hypertrophy and remodeling followed Wolff's law [18, 22, 23] and was serendipitously displayed due to the availability of limited number of sizes. When the implant was primarily attached proximally, as a consequence of the implant being either oversized or not seated firmly on the end of the transected bone, distal bone resorption occurred. Of particular interest was the anatomic change from distal metaphyseal cancellous bone to a configuration of cortical bone when proximal stress shielding occurred. This change was clearly demonstrated with SEM imaging at necropsy (Figs. 6, 7). It is known that when a bone is loaded below the modeling threshold range, disuse-mode remodeling is turned on and bone resorption would be observed with longer implant in situ time. When the bone is loaded above the modeling threshold value, remodeling would allow bone hypertrophy [8] to occur. Here, changes in cortical index between Time 0 and 12 months were used as a measure for quantifying the bone hypertrophy and resorption. The data indicated (Fig. 5) that direct distal bone loading, in general, allowed bone hypertrophy to occur. Furthermore, the cortical index measurement seemed to be sensitive in detecting cortical wall thinning associated with the stress shielding related bone resorption. It was therefore believed that cortical index measurements could serve as a valuable index for quantifying bone hypertrophy or resorption at specific regions in future studies.

The term osseointegration as initially defined by Per-Ingvar Branemark is the attachment of lamellar bone to implants without intervening fibrous tissue [1]. Thus, to validate the radiographic evidence of osseointegration, representative samples were subjected to histologic evaluation. The histologic data indicated that the distal bone tissues at the porous-coated regions were indeed in apposition with the periprosthetic (both cancellous and cortical) bone tissues, which had also grown into the porous-structured titanium without any interposing fibrous capsule. In six animals, where the implant was preferentially bound to the proximal fluted regions, remodeling had led to fibrous capsule formation within the smooth regions and marked bone adaptations within the distal porous-coated regions (Fig. 9F).

In conclusion, the radiographic and histologic results of this study appeared to confirm precise fitting of this distally porous-coated POP with a collar attachment, as a design principle, for the conservation of host bone tissue while allowing robust osseointegration to occur. Future research might correlate radiographic signs with histologic and mechanical parameters to create a grading scale for osseointegration, which could have a similar clinical relevance to total joint arthroplasty grading systems [4, 6, 10, 19]. Clinical applications of this approach might include assessment of whether implant design intentions are being met in animal translational studies and human preclinical

trials. Such longitudinal radiographic data could aid in the safe development of this much needed POP technology.

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