Sealing effect of hydroxyapatite coating on peri-implant migration of particles

AN EXPERIMENTAL STUDY IN DOGS

O. Rahbek, S. Overgaard, M. Lind, K. Bendix, C. Bünger, K. Søballe
From Aarhus University Hospital, Aarhus, Denmark

We have studied the beneficial effects of a hydroxyapatite (HA) coating on the prevention of the migration of wear debris along the implant-bone interface. We implanted a loaded HA-coated implant and a non-coated grit-blasted titanium alloy (Ti) implant in each distal femoral condyle of eight Labrador dogs. The test implant was surrounded by a gap communicating with the joint space and allowing access of joint fluid to the implant-bone interface. We injected polyethylene (PE) particles into the right knee three weeks after surgery and repeated this weekly for the following five weeks. The left knee received sham injections. The animals were killed eight weeks after surgery. Specimens from the implant-bone interface were examined under plain and polarised light.

Only a few particles were found around HA-coated implants, but around Ti implants there was a large amount of particles. HA-coated implants had approximately 35% bone ingrowth, whereas Ti implants had virtually no bone ingrowth and were surrounded by a fibrous membrane.

Our findings suggest that HA coating of implants is able to inhibit peri-implant migration of PE particles by creating a seal of tightly-bonded bone on the surface of the implant.

Received 18 October 1999; Accepted after revision 31 January 2000

Aseptic loosening is responsible for almost 80% of all revisions of total hip replacements (THR). Periprosthetic bone loss is thought to be caused by the generation of wear debris from the articulating surfaces and the bone-implant interface in both cemented and non-cemented THR. The biological response involves the recruitment of macrophages and the release of cytokines (interleukin-1β, interleukin-6), prostaglandin E2 and monocyte activating and chemotactic factors. The first experimental study to demonstrate the relationship between the accumulation of polyethylene (PE) wear particles and subsequent osteolysis in the interface was performed on rats. Since then, only a few studies have been able to produce osteolysis with wear particles in a stable interface, and the role of micromovement and implant loading in the pathogenesis remain to be investigated further in controlled, experimental studies.

Different ways of reducing rates of failure have been proposed. First, decreasing the production of wear particles by improving the quality of ultra-high-molecular-weight polyethylene is important. Secondly, several authors have suggested the possibility of a pharmacological intervention and lately bisphosphonates have shown promising effects in a canine model of total THR. Thirdly, inhibition of the migration of wear debris has been proposed. Wear particles are not only found in the joint, but also in the implant-bone interface. This suggests that the synovial cavity is in continuity with this interface, a concept termed "the effective joint space", which includes all periprosthetic regions that are accessible to joint fluid and thus to particulate debris.

The coating of implant surfaces influences the accumulation of wear particles in the implant-bone interface. In particular, the fibrous membrane surrounding uncemented smooth implants seems to provide a pathway for the migration of wear debris. By contrast, a porous coating has been shown to have a sealing effect as compared with a smooth implant surface both in experimental studies and also in the clinical setting. A similar effect of hydroxyapatite (HA) coating inserted in a press fit has been reported.

The unsolved problems regarding aseptic loosening and the disappointing results of conventional cemented prostheses after revision surgery and primary surgery in young-
er active patients have led to increased interest in cementless fixation.\textsuperscript{1,31} Clinically, the use of HA-coated femoral and acetabular components has shown promising short-term results.\textsuperscript{32,33} Controlled studies using roentgen stereophotogrammetric analysis have demonstrated that an HA-coated component gives significantly less early migration than a cemented component.\textsuperscript{33-37} Moreover, in animal studies HA has been shown to enhance bone growth across a peri-implant gap in both stable and unstable mechanical conditions.\textsuperscript{38} It is also able to convert movement-induced fibrous membranes surrounding unstable implants into bone.\textsuperscript{39-41}

Our aim was to investigate whether migration of PE particles was more likely to occur along the implant surface of a non-HA-coated implant than in an HA-coated implant. In addition, the migration of PE particles to distant organs and regional lymph nodes was studied histologically.

Materials and Methods

Design of the study. We used eight mature Labrador dogs with a mean weight of 26 kg (22 to 29). They had been bred for scientific purposes and were handled according to the Danish law on animal experimentation. We allocated randomly a loaded HA-coated implant and a Ti implant to each distal femoral condyle. If an HA-coated implant was allocated to the right lateral femoral condyle, then one was also placed in the left lateral condyle (Fig. 1). The test implants were surrounded by a gap which communicated with the joint space, allowing access of joint fluid to the bone-implant interface (Fig. 2).

Injection procedure. Three weeks after surgery a 5 ml solution of PE particles dispersed in sterile hyaluronic acid was injected into the right knee (Fig. 1). The left knee served as a control and received only hyaluronic acid. The injections were repeated weekly over a five-week period.

Fig. 1

Diagram showing the design of the study. An HA-coated (HA) and a non-HA-coated implant (Ti) were randomly allocated to either the medial or lateral condyle. Three weeks after surgery PE particles dispersed in sterile hyaluronic acid were injected into the right knee. The left knee served as control and received only hyaluronic acid. The injections were repeated weekly over a five-week period.

Characteristics of the implants. The implants were cylindrical (height, 10 mm; diameter, 6 mm) and made of titanium alloy (Ti6Al4V) with a grit-blasted surface. The HA coating was plasma sprayed (thickness, 50 \( \mu \)m; crystallinity, 68%; purity, 99%). The mean roughness (Ra) of the grit-blasted titanium surface was 1.12 ± 0.04 (SD) \( \mu \)m and of the HA-coated surface 1.25 ± 0.05 \( \mu \)m. The implants were fixed by a modified loaded Søballe\textsuperscript{39} implant device (Fig. 2) and had been sterilised by gamma irradiation.

Characteristics of the injected material. The polyethylene powder consisted of 100% pure crystalline high-density polyethylene (HDPE) (manufacturer’s information). The distribution of the particle size was determined by SEM (Cambridge S360; Cambridge Instruments, UK) with automatic image-analysis. The mean equivalence circle diameter was 2.09 \( \mu \)m (0.2 to 11) and the shape was spherical (Fig. 3). The powder consisted of 7% particles with a diameter below 1 \( \mu \)m.

The particles were gamma-sterilised. Immediately before use they were suspended in sterile hyaluronic acid (1.75 mg hyaluronic acid/ml phosphate-buffered saline, pH 7.4) and placed in an ultrasound bath for 30 minutes to homogenise the suspension. The final suspension contained 5 mg of HDPE (approximately \( 1.2 \times 10^9 \) particles) per ml of hyaluronic acid.

Operative technique. The implants were inserted using a sterile technique while the dogs were under general inhalatory anaesthesia. One implant was placed in each femoral condyle with a 0.75 mm gap to trabecular bone. The gap communicated with the joint space. HA-coated and Ti implants were randomly allocated to the medial or lateral
condyle for each dog (Fig. 1). An anterior parapatellar approach and medial arthrotomy were used. Vastus medialis and the patella were dislocated laterally. To avoid thermal trauma to the bone hand drilling was used to create a 7.5 mm hole leaving a 0.75 mm gap around the implants. The cavity was cleaned of bone debris with physiological saline water. After an anchoring screw had been inserted, the implant and polyethylene plug were mounted on the piston. The polyethylene plug protruded slightly above the cartilage so that a load was transferred through the implant system at each gait cycle (Fig. 2). Prophylactic antibiotics (Anhypen; Gis-Brocades, Holland) were administered immediately before and after surgery and analgesics (Temgesic, Schering-Plough Europe, Belgium; Paracetamol, Nycomed A/S, Denmark) were given daily for six days. Unrestricted weight-bearing was allowed after operation. The dogs were killed after eight weeks and the femora were immediately removed for histological analysis and biopsies of the synovia in both knees, the iliac medial lymph nodes, and the liver, spleen and lung were obtained for histological examination. Bacterial cultures were taken from all the knees.

Preparation of the specimens. The femora were cleaned of soft tissue and stored at -20°C. The specimens were machined using a water-cooled diamond blade (Exact Appartebau, Norderstedt, Germany). Each bone-implant specimen was cut in half parallel to the long axis of the implant at each gait cycle (Fig. 2). Prophylactic antibiotics (Anhypen; Gis-Brocades, Holland) were administered immediately before and after surgery and analgesics (Temgesic, Schering-Plough Europe, Belgium; Paracetamol, Nycomed A/S, Denmark) were given daily for six days. Unrestricted weight-bearing was allowed after operation. The dogs were killed after eight weeks and the femora were immediately removed for histological analysis and biopsies of the synovia in both knees, the iliac medial lymph nodes, and the liver, spleen and lung were obtained for histological examination. Bacterial cultures were taken from all the knees.

Evaluation

Decalcified sections and biopsies. These were stained with Oil Red O (ORO) and haematoxylin and eosin and examined under polarised and conventional light microscopy to evaluate the presence of PE particles and the histological response. Every third section produced was examined and a mean of sections (five to seven) was examined per implant.

Undecalcified sections. A stereological software program was applied (CAST-Grid; Olympus, Denmark A/S) for histomorphometry. This is based on user-specified grid counting on microscopic fields captured on the computer screen. With the use of vertical sectioning and the applied grid systems it was possible to calculate unbiased estimates using stereological methods assuming that there was no difference between the lateral and medial halves of the specimens. Every second section produced was analysed. A mean of five sections (5 to 7) was analysed per implant with a screen magnification of ×100 (attached to a light microscope with magnification of ×40). Ingrowth was defined as the surface of the implant covered by bone, bone marrow or fibrous tissue in percentage and was estimated using the linear intercept technique (approximately 350 intersections per implant). The gap volume was defined as the percentage of the initial peri-implant gap consisting of bone, bone marrow or fibrous tissue and was estimated using a point-counting technique (approximately 1750 points per implant). The initial gap area from the implant surface to 610 μm from the surface was counted.

Statistical analysis. The data are given as the mean and SD. Significance was determined by a paired or unpaired t-test. p values of less than 0.05 (two-tailed) were considered to be significant.

on a newly developed microtome (KDG-95; Ignition, Bilthoven, The Netherlands) with 350 μm between sections. They were counterstained with 4% Light Green.
All the dogs completed the study and walked without limping one to two weeks after surgery. In one the wound was resutured after evacuation of a subcutaneous hematoma. Bacterial cultures from eight of the knees showed scattered growth of *Staphylococcus albus* probably because of contamination during sampling, since no clinical signs of infection were noticed.

When preparing the decalcified specimens, the surfaces of the implant were easily separated from the surrounding tissue except for one Ti implant from a PE-particle-exposed knee in which the interface was lost.

**TI implants** (Figs 4 and 5). A thin fibrous membrane with a lining of synovial-like cells surrounded all the Ti implants. The layer beneath the synovial lining was rich in extracellular matrix with fibres orientated parallel to the surface of the implant. In this layer scattered spindle-shaped cells predominated. The deep layer of the membrane close to the bone was rich in capillaries and larger vessels. Small islands of fibrocartilage were occasionally found, especially in the luminal part (close to the joint space) of the membrane. Large amounts of PE particles were seen along the entire length of Ti implants in the PE-particle-injected knees. No signs of inflammation were present and the membranes resembled those around implants from sham-injected knees. No formation of foreign-body giant cells was seen.

**HA implants** (Figs 4 and 5). These were all surrounded by bone marrow with growth of trabecular bone into the HA coating. Synovial lining cells were not present along the surface of the implant. Occasionally, a few scattered particles were found in the marrow near the joint space in the PE-particle-exposed knees. Around one implant some particles were found in the basal part opposite the joint cavity. No inflammatory reaction was seen.

**Synovial biopsies.** Histological examination showed no signs of infection. In the particle-exposed knees PE particles were located beneath the synovial lining cells along with lymphocytes and plasma cells. Multinucleated for-
Table I. Histomorphometrical data (mean percentage; SD) on ingrowth and gap healing in bone and fibrous tissue (+PEP = implants from particle-injected knees)

<table>
<thead>
<tr>
<th></th>
<th>HA-implants/ control</th>
<th>HA-implants/+PEP</th>
<th>TI-implants/ control</th>
<th>TI-implants/+PEP</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Ingrowth</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Bone</td>
<td>35.1 (8.9)</td>
<td>37.1 (6.5)</td>
<td>0.5 (0.8)</td>
<td>1.1 (2.1)</td>
</tr>
<tr>
<td>p &lt; 0.01</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Fibrous tissue</td>
<td>9.3 (8.4)</td>
<td>NS</td>
<td>2.4 (5.5)</td>
<td>90.4 (16.3)</td>
</tr>
<tr>
<td>p &lt; 0.01</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Gap healing</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Bone</td>
<td>22.1 (6.7)</td>
<td>28.5 (8.3)</td>
<td>11.5 (3.9)</td>
<td>14.1 (7.5)</td>
</tr>
<tr>
<td>p &lt; 0.01</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Fibrous tissue</td>
<td>6.6 (4.8)</td>
<td>NS</td>
<td>3.5 (4.7)</td>
<td>67.1 (17.1)</td>
</tr>
<tr>
<td>p &lt; 0.01</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

eign-body giant cells surrounded some of the particles. In the synovium from one dog a nodular collection of lymphocytes was found. Synovium from the control knees appeared to be normal.

**Regional lymph nodes.** Histological examination of the right-sided medial iliac lymph nodes by polarised light and ORO revealed many particles distributed both in the cortex and the medullary area, whereas none was seen on the left side.

**Distant organs.** Biopsies from the spleen and liver showed no particles. Some birefringent particles could be seen in the lung tissue, but these did not stain with ORO.

**Histomorphometry.** No significant differences in ingrowth and gap healing were found between implants from control and PE-particle-injected knees (Table I). In both groups, however, there was a huge difference in both ingrowth and gap healing between Ti implants and HA-coated implants. The latter had in both groups approximately a 35-fold increase in bone ingrowth as compared with non-coated implants, which were almost completely covered by fibrous tissue (approximately 90%).

**Discussion**

Our study has shown that HA-coated implants are able to inhibit the migration of PE particles in the interface by increased bone ingrowth to the surface of the implant. By contrast, the thin fibrous membrane surrounding the non-coated implants contained a huge number of PE particles. The effect of bony anchorage of the HA-coated implants on the migration of PE particles can be explained by the fact that it acts either as a mechanical barrier or that it increases the stability of the implant. The transport of wear particles by the pumping of fluid in the unstable interface can...
therefore be avoided. A difference in the roughness of the surface of the implant may influence the migration of PE, but it seems unlikely that a slight difference in the roughness of HA-coated and non-coated implants would influence the migration of particles in a 0.75 mm gap.

In our study, the results confirm earlier findings, namely that HA enhances the ingrowth of bone across a gap, which yields a superior initial fixation as compared with non-HA implants. The gap model developed for this study seems to be clinically relevant, since cementless prostheses carefully inserted in a press fit will have areas with gaps between the implant and the bone. This problem must be considered to be even greater in revision surgery of cemented arthroplasties or in patients with bone defects or osteopenic bone. Gaps between the implant and bone must be regarded as an extension of ‘the effective joint space’,23 and therefore as a potential pathway for the migration of wear debris. Along with earlier studies,26,28,30 our study shows that it is important to fill the interface with bone instead of fibrous tissue and for this purpose the use of an HA-coating seems promising. HA-coated implants had approximately 35-fold more bone ingrowth and a larger bone volume in the initial gap as compared with non-HA-coated implants.

In previous studies, the porous implant surface has been shown to have a sealing effect when inserted in a press fit,28,36 but to our knowledge the effect of a porous coating on the migration of particles inserted in non-interference fit remains to be investigated. Based on our present and previous studies,39,44 it can be speculated that porous-coated implants with an HA coating will have a superior effect on the migration of particles in a gap as compared with non-HA-porous-coated implants, because of early bony anchorage.

Howie et al12 first studied the influence of PE particles around implants in an animal model. Their model created a cement-bone interface by inserting a non-loaded acrylic plug into the distal femur of rats. Resorption of bone in the cement-bone interface was induced after only eight weeks of repeated intra-articular injections of powdered HDPE. In our study PE particles had no effect on tissue ingrowth or gap healing around implants. Furthermore, the PE particles in the fibrous membrane around Ti implants did not seem to alter the morphology of the membrane and no cystic cavities were found in the interface as previously described by Howie et al.12 A recent study, however, using many different types of particle in Howie’s model could not reproduce the original finding of bone resorption.15 The dosage of particles used in our study was based on an estimated weekly production of 9.6×10⁷ particles in human total hip replacements.26 Assuming that the effective joint space in a human hip has a volume of approximately 45 ml, the weekly load was calculated to be 2×10⁸ particles per ml. By injecting 6×10⁹ particles per week into the knee of dogs (assumed volume, 15 ml), we exposed the joint cavity to twice as high a load as in the human hip, namely 4×10⁹ particles per ml joint. This was done to accelerate the inflammatory process.

The mean size of the particles was larger than that of particles isolated from periprosthetic tissue in retrieval studies.46,47 An inflammatory response has been shown, however, after only six weeks of stimulation with HDPE particles of approximately the same mean size (2.03 µm).13 In terms of peri-implant migration it can be speculated that the HA coating is less effective on smaller than on larger particles. This would be true if the bony fixation to the implant only acts as a sieve and therefore only as a mechanical barrier to larger particles. We believe that it also prevents the movement of joint fluid, which carries the particulate debris to the interface, thus reducing the migration of particles in all sizes. Further studies, however, are needed to clarify this.

A recent study by Aspenberg and Herbertsson14 has questioned the osteolytic effects of HDPE particles when applied to an implant interface, but demonstrated that the particles could maintain a peri-implant membrane induced by movement. The model used in that study, however, lacked the influence of an inflamed joint and the particles were applied to cortical bone instead of trabecular bone. We find our experimental implant device model unique since it is loaded, the joint is inflamed, and the implant device allows access of joint fluid to the bone-implant interface, thus mimicking the clinical situation with the ‘effective joint space’. Aspenberg and Herbertsson14 suggested that mechanical stimuli are of primary importance for prosthetic loosening and the effect of wear debris intervenes at a later stage. Until now, there have been only a few reports of the combined effect of implant movement and PE particles, but there seems to be increasing evidence that it is the combined effect of micromovement and particulate debris which causes the aggressive membrane around loose implants.15,48,49

Our findings suggest that HA coating of implants is able to inhibit peri-implant migration of PE particles by creating a seal of enhanced bone ingrowth. Further studies are in progress to investigate the long-term effect of the HA coating on the loosening of implants.

We thank Anette Milton and Jane Pauli for technical assistance. Biomet Inc kindly delivered the implants and Smith and Nephew-Richards provided the particles. The study was financially supported by the Danish Rheumatism Association, the Danish Research Council, the University of Aarhus, Denmark and the Consul J. Fogh Nielsen and E. Fogh Nielsen’s Foundation.

One or more of the authors have received or will receive benefits for personal or professional use from a commercial party related directly or indirectly to the subject of this article.

References


