Histological evaluation of allograft incorporation after cemented and non-cemented hip arthroplasty in the goat

3 Histological Evaluation of Allograft Incorporation after Cemented and Non-cemented Hip Arthroplasty in the Goat

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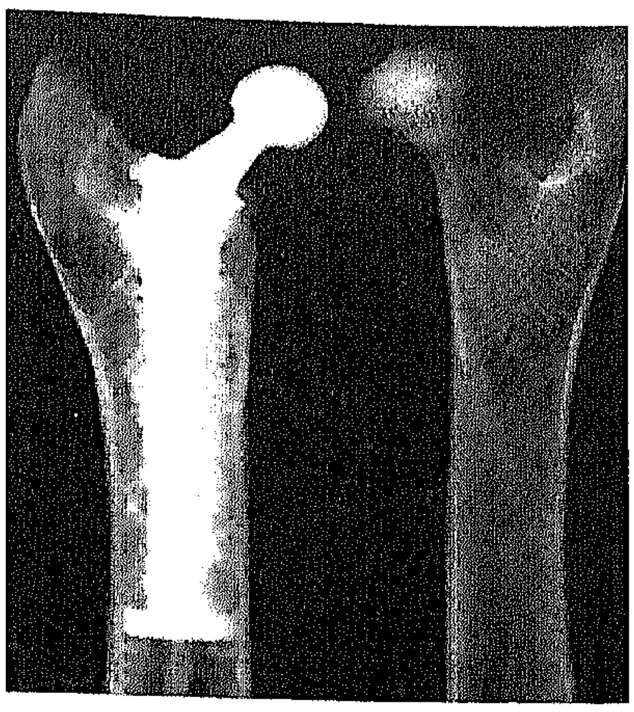
One of the major complications associated with the very successful operation of artificial hip replacement is the bone stock loss that may be found after aseptic loosening of the prosthesis. This complication hampers the successful fixation of a new prosthesis. In the past, orthopaedic surgeons tried to resolve this problem by the use of revision prostheses with longer stems and/or reinsertion of a standard prosthesis with more cement. However, these procedures are not very elegant, since in a second revision procedure further bone stock deficiencies will be found, and the problems for the orthopaedic surgeon will be increased.

The challenge to the orthopaedic surgeon is to restore the original bone stock during revision surgery. This goal can only be reached if the lost bone stock is supplemented with graft material, which has to be subsequently incorporated into a new bony structure. Different types of bone may be used in the grafting procedure: cortical bone, cancellous bone, chips or larger intact pieces of bone, fresh or deep frozen and lyophilised bone, and autograft or allograft bone (Oikarinen and Korhonen 1979, Mellonig et al. 1981, Kohler et al. 1986, Kakaiya and Jackson 1990, Alho et al. 1989, Wilson et al. 1989).

We present the first results of a study in the goat which investigated the incorporation of graft in combination with cemented and non-cemented prostheses. In our orthopaedic department, chip-like trabecular bone grafts were previously used with good results to restore the bone defects associated with acetabular protrusion (Slooff et al. 1984). The present study was designed to evaluate the feasibility of femoral revision surgery with morsellised allograft chips. Allograft bone was harvested from the sternum of donor goats. A straight stem cemented prosthesis or an experimental hydroxyapatite-coated titanium non-cemented (Osteonics) prosthesis was used. The operation technique and the first biomechanical results are described in Chapter 18.

Methods and Materials

In order to visualise new bone formation, the goats were labelled with different types of fluorochrome during the study. The animals were anaesthetised and fixed by perfusion. Thick sections of the femora were made with the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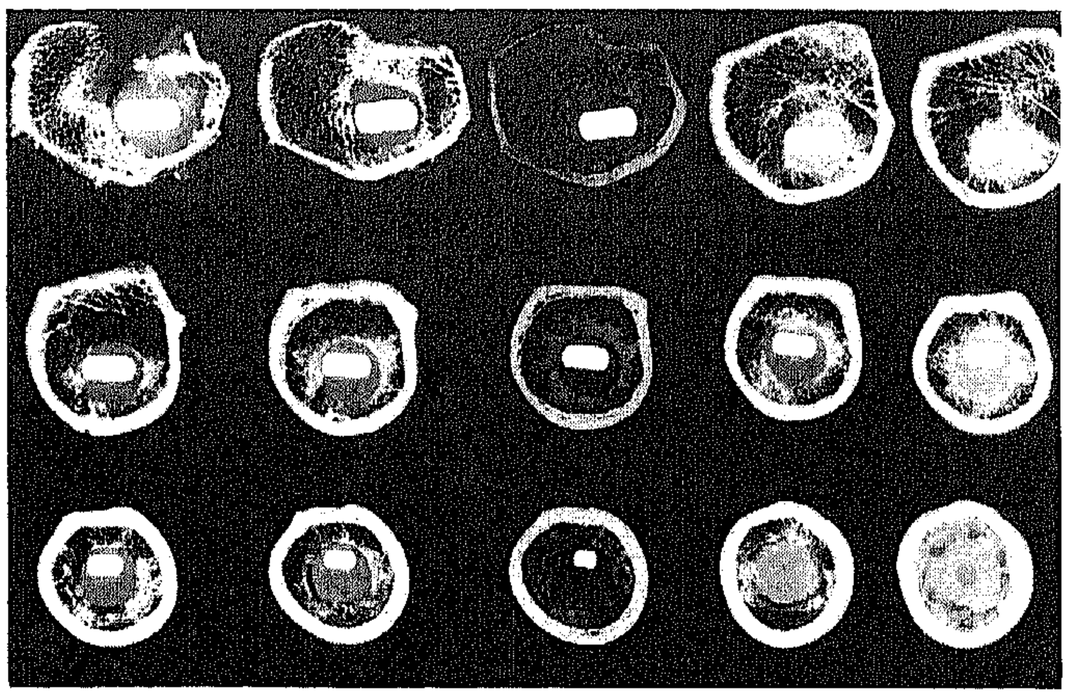
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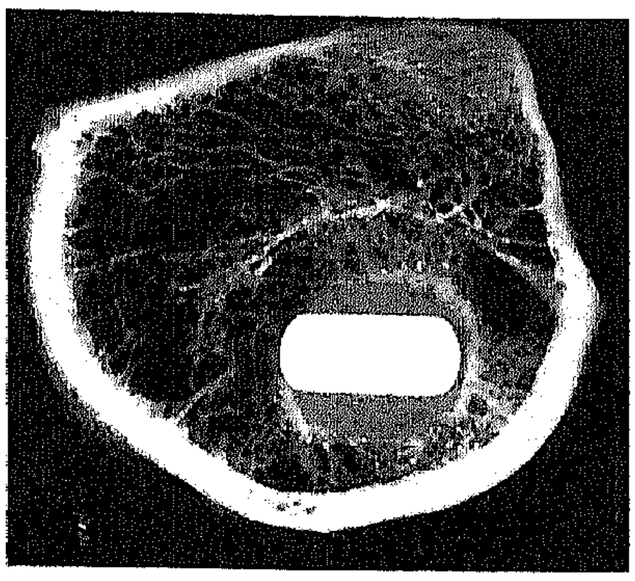
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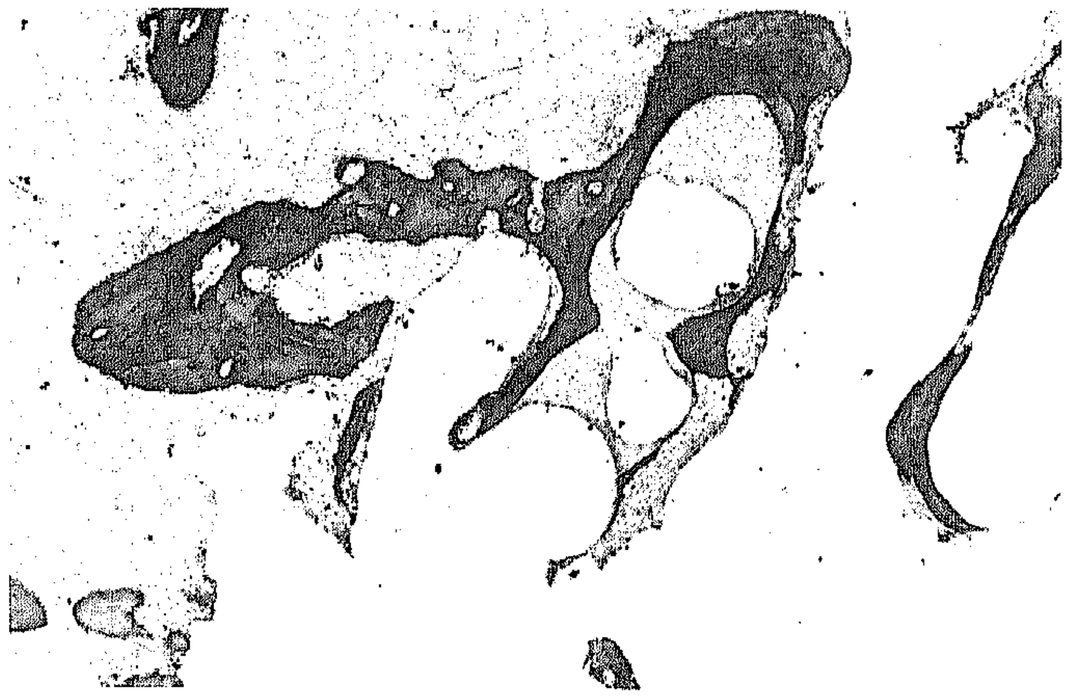
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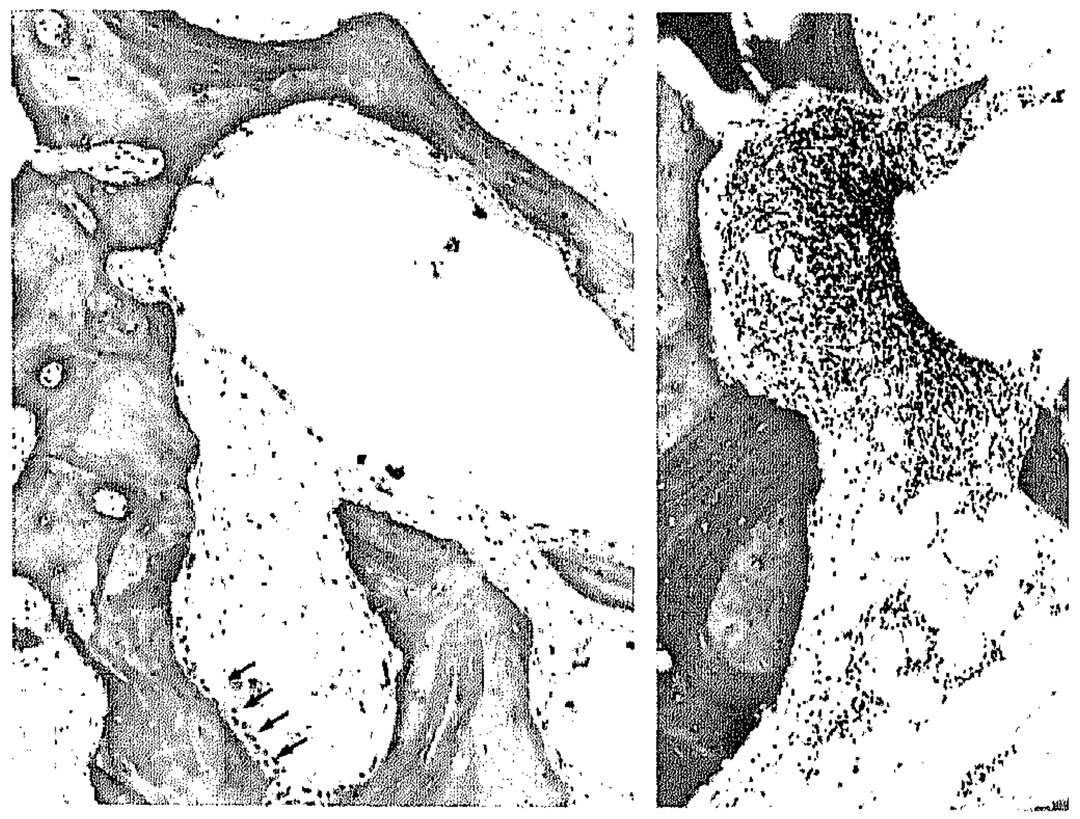
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Fig. 3.1. a Roentgenograph of cemented prosthesis with intramedullary augmentation with allograft chips (×0.55). **b** Roentgenograph of thick sections from proximal (upper row, left) to distal levels (lower row, right) (×0.65). **c** Enlargement of fourth section shown in b. Note trabecular structure between the cement layer and the pre-existing trabecular bone (×1.7). **d** Low magnification haematoxylin–eosin stained section of mid-shaft level. Note trabecular system that connects the cement layer with the pre-existing compact cortical host bone (×18). **e** Interlock between cement and graft. The cement was resolved during histological preparation of the section but could be recognised by the barium sulphate (×35). **f** Detail of interlock between cement and graft. Dark areas in the bone are graft, with empty osteocyte lacunae, the lighter areas represent new bone. Note thin soft tissue interface between cement and graft, and area of active bone apposition (arrows) (×90). **g** Local accumulation of lymphocytes (×80).

prosthesis in situ. Rocntgenographs of the sections were made, and the surfaces of the sections were stained with basic fuchsin. For the first evaluation of graft incorporation we applied fluorescence microscopy at low magnification. For the visualisation of the hydroxyapatite interface between prosthesis and new bone we used a Biorad confocal microscope. Subsequently, the sections were prepared for further histology. Undecalcified sections were stained with basic fuchsin and embedded in methylmethacrylate; thin sections were stained with haematoxylin–eosin. In the histological analysis, special attention was paid to graft lysis, incorporation of the graft, localisation of osteoclasts and osteoblasts, contact area of bone with cement and hydroxyapatite, the presence and extent of the periosteal reaction, immunological reactions to the graft, vascular invasion of the graft and the presence of infections.

Operations were performed on 12 goats; six received a cemented prosthesis and six a non-cemented prosthesis. Three goats in each group were sacrificed after six weeks and three after 12 weeks. Since not all histology was completed, only the results of the first two specimens can be presented here. Three specimens showed clear signs of infection after histological analysis: two cemented prostheses and one in the non-cemented group. Infected prostheses showed large numbers of polymorphonuclear leucocytes, lysis of the graft and bone loss. All infected goats were from the six week groups. These infected specimens were not analysed further so that of the six week cemented arthroplasties, only one remained for histological analysis. Therefore this chapter will focus on the results that were found after 12 weeks.

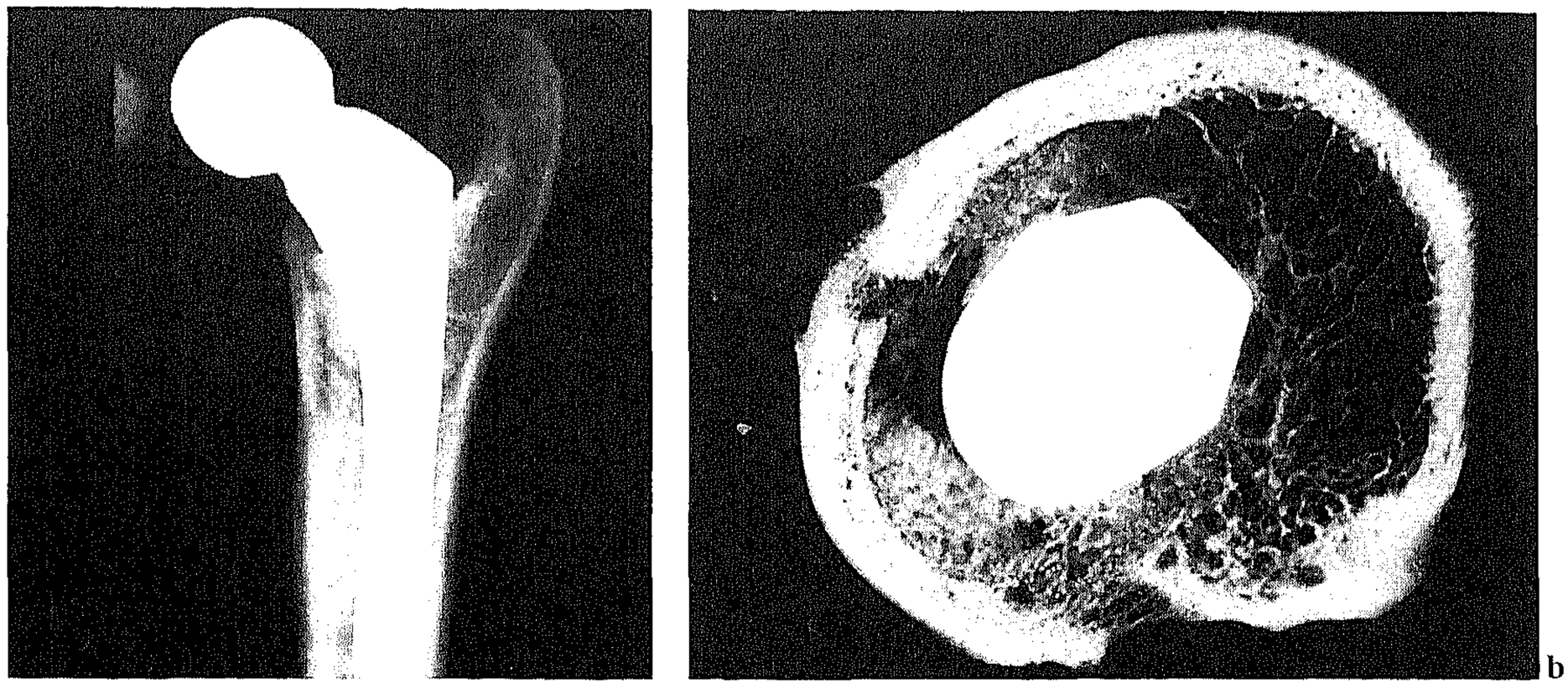
Cemented Arthroplasty

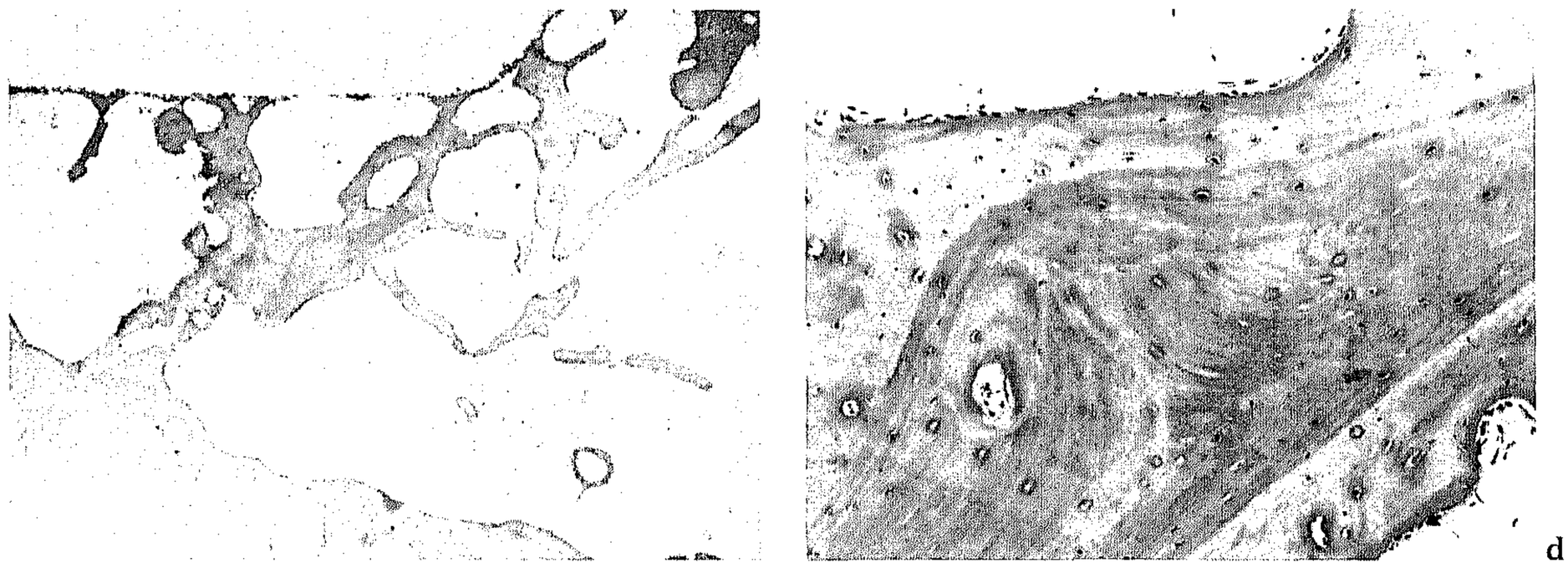
The sections showed that the prosthesis was surrounded by a homogeneous cement mantle and a layer of impacted pieces of trabecular bone graft 3mm thick. In the impacted graft, small pieces of original trabecular bone could be recognised. The spaces in between the graft were initially filled with a fibrin clot. After incorporation of the graft, there was a very good interlock between the pre-existing trabecular bone, the graft and the cement, as if a new trabecular structure had been formed (Fig. 3.1b,c). The basic fuchsin and the fluorescence microscopy (at low magnification) showed a trabecular structure in close contact with the cement mantle. Calcein green fluorescence labelling indicated that there was active bone turnover before sacrifice of the animal. At more distal levels both the roentgenographs and the fluorescence microscopy again showed that a good interlock existed between the cement, the graft and the compact cortical bone (Fig. 3.1b). Indeed, histology at low magnification showed trabecular bone interconnecting the cortical bone with the cement mantle (Fig. 3.1d). Although the cement was resolved during preparation of the histology, remnants of the barium sulphate remained in the sections. At all levels studied, the cement had penetrated into the graft (Fig. 3.1e,f). At high magnification, the original graft could be recognised by the empty osteocyte lacunae (Fig. 3.1g,d). It appeared that the trabecular bone was a mixture of graft and new bone which was characterised by the filled osteocyte lacunae. Rows of osteoblasts indicated that active bone formation took place (Fig. 3.1f). Changing numbers of lymphocytes were always seen in the graft, both with the cemented and non-cemented prostheses (Fig. 3.1g). No bone destruction was apparent in the direct vicinity of these cells which were probably a result of a mild immunological reaction to the allograft.

Occasionally, a relatively large protrusion of the cement mantle was seen in contact with the cortical bone. Locally, the new bone was contacting the cement mantle without a soft tissue interface. However, a thin soft tissue interface was generally present between the cement layer and the graft. Also macrophages were found at the interface between cement and graft indicating an immunological reaction to the cement mantle. The fluorescence microscopy at higher magnification showed active bone formation in the direct vicinity of the cement mantle.

The Non-cemented Arthroplasty

The roentgenograph of the femur after 12 weeks showed that the tip of the prosthesis was in very close contact with the cortex of the femur (Fig. 3.2a). Roentgenographs of the thick sections showed a very intimate contact between the graft and the prosthesis, particularly at the proximal levels (Fig. 3.2b). Fluorescence microscopy and basic fuchsin stained sections showed that the calcein green had been incorporated into the





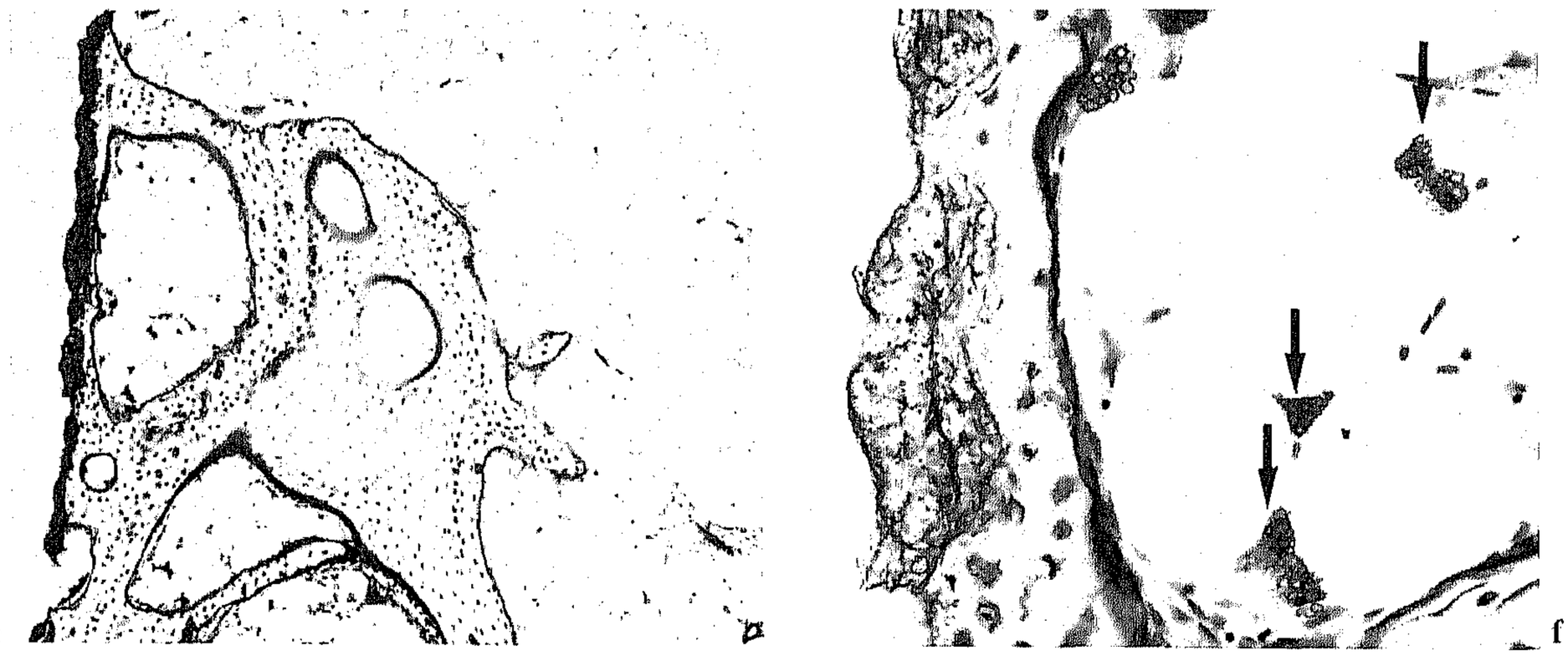


Fig. 3.2. a Roentgenograph of non-cemented prosthesis after insertion in the femur (×0.55). **b** Roentgenograph of thick section through proximal part of femur. Note very intimate contact between the trabecular bone and the corners of the prosthesis (×1.7). **c** Low magnification haematoxylin-eosin stained proximal section through lateral part of the femur. Note intimate contact between trabecular structure and the corner of the prosthesis (×18). **d** Detail of c, showing that the trabecular bone is a mixture of graft (empty osteocyte lacunae, darker stained bone) and new bone (filled osteocyte lacunae) (×120) **e** Interlock between the layer of hydroxyapatite and bone (×64). **f** Detail of **e**. Note isolated thydroxyapatite crystals between the medullary fat cells (arrows), and very intimate contact of new bone with the hydroxyapatite layer (×300).

trabecular bone. The new bone appeared to be in very close contact with the prosthesis (Fig. 3.2c,e,f). Confocal microscopy confirmed that there was a very good interlock between the new bone, the hydroxyapatite layer and the prosthesis. After removal of the prosthesis, this layer of hydroxyapatite stuck to the newly formed bone, which was present in between the layer of hydroxyapatite and the graft (Fig. 3.2e,f). With fluorescence microscopy of the same area, active bone apposition was still seen. Trabeculae connected the prosthesis with the host cortical bone, particularly at the lateral side of the prosthesis (Fig. 3.2c). At high magnification it became clear that this trabecular bone was again a mixture of graft and newly formed bone (Fig. 3.2d).

Roentgenographs of thick sections indicated that at midshaft and distal levels the graft was not in contact with the prosthesis and cortical bone. Ingrowth of new bone only appeared to have taken place in the region where the tip of the prosthesis was in close contact with the cortex of the bone. The graft had resolved and young trabecular woven bone was growing locally into the space between prosthesis and cortical bone. A bony bridge between prosthesis and cortical bone had only been formed where the tip of the prosthesis was in close contact with the lateral cortex of the femur.

Conclusions

The histological protocol used in the present study allowed a good evaluation of the graft incorporation. With this procedure of intramedullary augmentation with morsellised allograft chips a very good homogeneous wall of chips can be made. Signs of incorporation were found both in the graft in combination with the cemented arthroplasty, and in the graft around the non-cemented prosthesis.

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