Detection, prevention and direct post-operative intervention in orthopaedic implant infection

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Detection, prevention and direct post-operative intervention in orthopaedic implant infection

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Contents

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INTRODUCTION and AIM OF THIS THESIS

Introduction

In modern medicine, biomaterials are increasingly used to support or restore human body function. A biomaterial can be defined as man-made material designed to interact with living tissue or with body fluid. Joint prostheses, heart valves, external fixator pins, catheters, and contact lenses can all be considered examples of successful applications of biomaterials.
Despite the fact that biomaterials have led to great improvements in medicine, they all have one thing in common: they tend to attract infectious micro-organisms leading to the occurrence of biomaterial-associated infections. The biomaterial itself then has become the focus of infection. These infections are mainly caused by direct contamination during surgery, but they can also be caused by haematogenous spread of bacteria from an infection site somewhere else in the human body. The clinically important step of bacterial attachment to the surface of a biomaterial is then followed by aggregation of other bacteria and growing of the bacteria, resulting in biofilm formation.
Biomaterial-associated infections can cause severe problems, from malfunction of the biomaterial implant to lethal sepsis. Furthermore, treatment of these biomaterial-associated infections is complicated, as biofilms grow slowly (Costerton et al. 1999) and the micro-organisms involved are more resistant to antibiotics than their planktonic counterparts (Stewart and Costerton 2001).
The extent of the infection problems in orthopaedic implant surgery in all of its magnitude became clear to us at the Academisch Ziekenhuis, Groningen, the Netherlands after starting the infection complication registration in 1996. This was just one year after the orthopaedic-Operating Room (OR) moved from the secluded facilities in the "Vrouwen-Kliniek" to the main hospital building. With this change of facility, no specialized operating-room personnel specifically dedicated to

the orthopaedic specialty was available anymore and regular exchange of personnel hampered proper information of the team that performed biomaterial-associated surgery. As a consequence, the knowledge among OR-personnel about biomaterial-associated complications slowly diminished to a very dangerous level. At the onset of this study in the period 1999-2000, our orthopaedic ward had a Surgical Site Infection of 6.9%, which was above the national level. For Total Hip Prostheses, the total infection rate was 8.8%, with 3.7% deep infections. The total infection rate in revision surgery was 6.9%, with 3.4% deep infections (PREZIES registration November 2002). These unacceptable figures prompted us to investigate possible causes and evaluate different preventive measures. Collaboration already had been set up between the departments of Orthopaedic Surgery and BioMedical Engineering, UMCG, Groningen, where the control of biomaterial-related infections is a topic area. After the initial work of H. van de Belt (2001) en D. Neut (2003) one of the causes of these high infection numbers in revision surgery was thought to be the difficulties in diagnosing a low grade septically loosened total joint prosthesis.
As a starting point we assumed that during the primary procedure a larger part of the implanted total joint prostheses was contaminated. This assumption formed the base of our thought that aseptic loosening of total joint prostheses is more or less a myth.

Aim of this thesis

The aim of this thesis is therefore to investigate the reasons for diagnostic problems of infection in total joint prostheses in a University Hospital setting. Diagnostic problems are analyzed during the work up for revision surgery as well as during the peri-operative hospital stay after primary hip replacement. A

possible method of preventing clinical signs of infection of a percutaneous orthopaedic implant, which is even more susceptible to infection than totally internal implants, is investigated in an animal model.

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BIOMATERIAL-ASSOCIATED SURGERY AND INFECTION
A review of literature

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Introduction

The incidence of wound infection after clean surgery is often underestimated. Infection rates up to 15% can be found by meticulous follow up (Leaper and Melling 2001). The consequences of these complications can be troublesome for the patient involved. Most of the time the post-operative recovery will be delayed and secondary healing of the operative wound will occur. The long term consequences of the infection will mostly be within acceptable limits. When biomaterials are involved however in post-operative infectious complications, a totally different scenario is likely to occur and the longevity of the artificial organs and temporary assist devices is limited. Biomaterial-associated infections are usually resistant to antibiotics and removal of an infected implant is the final outcome of most of these infections at high costs for the health-care system and discomfort for the patient. Ever since the description by Gristina of biomaterial-associated infection as "a race for the surface" (Gristina 1987, Gristina et al. 1988[a]) between microbial adhesion and tissue integration, there is a growing awareness of the risk of foreign body implantation. The design of a biomaterial surface upon which the race for the surface is fought, determines the outcome of it, as it depends upon a delicate fine-tuning of the properties of the biomaterial surface that has not yet been achieved.

Some infected biomaterial implants are relatively easily removed, like contact lenses (Liesegang 1997), voice prostheses (Ackerstaff et al. 1999) or dentures (Radford et al. 1999). The total artificial heart (Gristina et al. 1988[b]), elongatable endoprostheses as used after extensive tumour resection in children, total hip and knee arthroplasties on the other hand are much more difficult to remove. Moreover, removal of these devices often constitutes a clinical dilemma, as for instance the removal of an infected Hickmann catheter in

patients on chemotherapeutic treatment. Here the surgeon has to choose between two evils: leaving the infected catheter in place or removal at the expense of stopping the chemotherapy (note that a new catheter can only be safely inserted once the infection has fully cleared, otherwise recurrence will happen in due time). Biomaterial implants sometimes are complex devices made of a combination of different biomaterials. These materials need to be compatible with their biological environment, which is not always the first concern of the biomedical engineer, as mechanical and manufacturing properties often dictate the choice for a given material.

Table 1. Incidences of infection of different biomedical devices in permanent contact with skin and/or outer human body environment.

Body site	**Implant or device**	**Incidence of infectious complications necessitating exchange**
Urethra	Foley catheter	2.8/1000 catheter days (Luehm and Fauerbach 1999)
Venous system	Peripheral inserted central venous catheters	2-5/1000 catheter days (Safdar and Maki 2005)
Arterial system	Arterial catheters	0.4-0.7% (Frezza and Mezgebe 1998)
Intraperitoneal	Peritoneal dialysis catheters	11-13% (Thodis et al. 2005)
Extremities	Pins in external fracture fixation	12-71% (Bernardo 2001)
Oral cavity	Dental implants	5-10 % (Ehrlich et al. 2005)
Laryngeal cavity	Voice prosthesis	Every 4 months (Van den Hoogen et al. 1996)

Tables 1 and 2 list commonly used biomedical implants in modern medicine with their incidence of clinical infections.

Table 2. Incidence of infection of different biomedical implants arranged according to body site.

Body site	**Implant or device**	**Incidence of infection**
Subcutaneous	Cardiac pacemaker	1-5% (Borer et al. 2004)
	Tissue expanders	0.9% (Disa et al. 1999)
	Chin augmentation implants	0.8% (Gross et al. 1999)
Soft tissue	Mammary prosthesis	2-2.5% (Pittet et al. 2005)
	Abdominal wall patches	3-8% (Deysine 1998)
	Penile prostheses	2-10% (Schoepen and Staerman 2002)
	Nasal implants	3.2% (Godin et al. 1999)
	Intraocular lenses	0.5% (Kahn et al. 2005)
Circulatory system	Prosthetic heart valve	1-3% (Ehrlich et al. 2005)
	Dacron aortoiliofemoral bypasses	2-10% (Andreev 1995)
Bone	Total Hip Arthroplasty	1% (Zimmerli et al. 2004)
	Total Knee Arthroplasty	2% (Zimmerli et al. 2004)

Different biomaterials are prone to infection by different organisms. *Staphylococcus aureus* is generally found on metallic implants (Barth et al. 1989), while pseudomonas and *Staphylococcus epidermidis* are mainly isolated from polymeric implants (Barth et al. 1989, Ferreiros et al. 1989). Consequently, as more different biomaterials are involved in an implant, this increases the chance of a biomaterial-associated

infection and the recognition of strains being pathogenic. *S. epidermidis* was long considered a non-pathogenic and harmless member of the normal skin micro flora, but only became a pathogen in the era of biomaterial-implants.
Surgery is supposed to be performed in a sterile way, but it can well be argued that completely sterile surgery is impossible. In a contamination study of primary total hip arthroplasties, 30% of the materials in contact with the prosthesis site harvested viable micro-organisms (Maathuis et al. 2005). Nearly the same percentage was found by Knobben et al. in two different studies (Knobben et al. 2006a,b).
Troublesome in biomaterial-associated infections is the long history of antibiotic therapy applied prior to the ultimate decision to remove the implant, giving the opportunity for antibiotic resistance to develop. Van de Belt et al. (1999) described the culturing of antibiotic resistant staphylococci from gentamicin-loaded bone cement that was removed in a hip revision for infection. The path of entry of infecting micro-organisms to a biomaterial implant can be directly along the parts of the implant itself, like along the polyvinylchloride drive lines of the total artificial heart (Gristina et al. 1988) or through haematogenous spreading (Sanderson 1991) or dental treatment (LaPorte et al. 1999). Alternatively, it can be stated that, despite the use of intra-operative systemic antibiotic prophylactics, strict hygienic protocols, sterile operating theatres and special sterile enclosure, the possibility exists that prostheses become contaminated during surgery and will be implanted in this state. Subsequently, whether or not clinical signs of infection develop depends on interplay of the host immune system and the microbiological characteristics of the infecting organisms.
In this chapter we present an overview of the mechanisms of biomaterial-associated infection and its occurrence in various medical disciplines. Surgical procedures are critically reviewed comparing non biomaterial-associated versus biomaterial-

associated surgery and recommendations are given for biomaterial-associated surgery.

The "race for the surface" and biofilm formation

Several authors have proposed a model for biofilm formation in general (Busscher et al. 1996, Van Loosdrecht et al. 1990), which has been developed from to the concept of "the race for the surface", as first formulated by Gristina in 1987.
Micro-organisms have a strong tendency to become attached to surfaces. On these surfaces they form a micro-ecosystem in which different microbial strains and species grow in a slime-enclosed biofilm. Biofilm formation involves a sequence of events (Busscher et al. 1996, Van Loosdrecht et al. 1990), represented in Figure 1. The first step is the adsorption of small, macromolecular components that form a so called "conditioning film" on the surface of the biomaterial involved.
The formation of this conditioning film is extremely fast and occurs in seconds after exposure to a biological environment. The biological environment in which the biomaterial is placed determines the nature of the adsorbed macromolecules. For instance, dental restorative materials adsorb salivary proteins; contact lenses adsorb proteins and lipid components from tear fluid, while blood contacting biomaterials adsorb a variety of different plasma proteins prior to the arrival of the first micro-organism. A prerequisite for microbial adhesion to occur is an adsorbed conditioning film, which changes the physico-chemical properties of the interacting surfaces. Adherence of micro-organisms on bare biomaterials surfaces is rare.
The initial adhesion of micro-organisms is reversible and depends on the overall physico-chemical characteristics of the microbial cell surface, the biomaterials surface and the biological bathing fluid. Firm anchoring through exopolymer production may change this reversible adhesion to an

irreversible state. The exopolymers surrounding the micro-organisms embed the biofilm to form the so-called "glycocalix" (Neu et al. 1992).

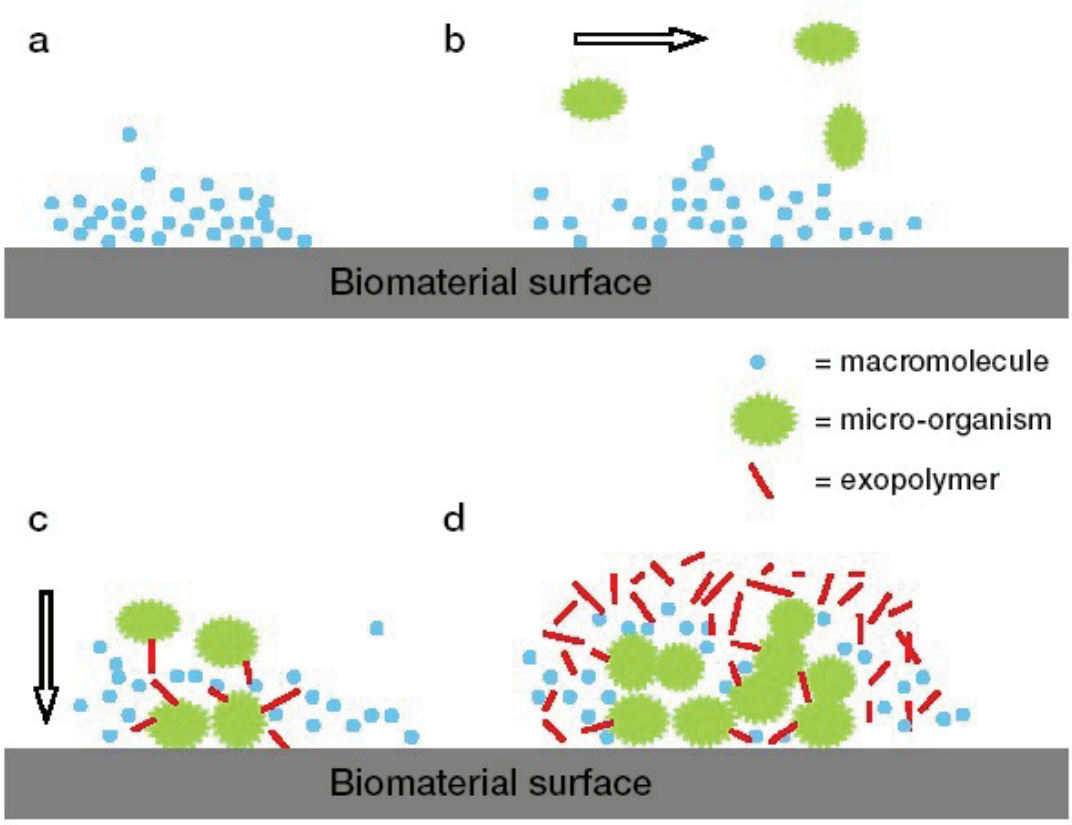

Figure 1. Sequential steps in the formation of biofilms on a biomaterial surface, including: Formation of a conditioning film (a). Microbial mass transport (b). Initial microbial adhesion and anchoring through exopolymer production (c). Growth of adhering micro-organisms (d).

In addition to anchoring, the glycocalix offers protection against environmental attacks and antibiotics (Isaklar et al. 1996, Schierholz and Beuth 2001, Sugarman and Young 1989). Multiplication of the adhering organisms is the main mechanism of growth in a biofilm and eventually leads to the formation of a thick film. The growth rate due to a lowered metabolism is generally slowed down in the biofilm as compared with a planktonic state of growth. Because of this lowered state of metabolism, the sensitivity for certain antibiotics is reduced.

Also, bacteria in this quiescent state are hard to detect with standard microbiologic techniques. This puts the concept of "aseptic loosening" in for example orthopaedic implant surgery in another perspective, as will be discussed later.
In the final phase of biofilm formation organisms on the periphery of the expanding biofilm may detach or disaggregate, which plays an important role in the pathogenesis of septic processes.

Biomaterials and micro-organisms

The host defence is significantly compromised in the presence of a foreign material (Elek and Conen 1957). In continuation of this concept the resistance of osteomyelitis and foreign body-related infections to antibiotic therapy was rationalized by others (Lam et al. 1980, Nickel et al. 1994). Furthermore, the relatively avirulent *S. epidermidis*, normally not capable of establishing infection, has become the most common causative organism in biomaterial-associated infection (Christensen et al. 1989).
The organisms causing a biomaterial-associated infection may have one or more of several sources. The first source is constituted by the skin. During insertion of the biomaterial, micro-organisms from the skin can be pushed towards the implant surface. A second source is constituted by airborne micro-organisms, which in varying concentrations are normally present in the operating theatre. They can reach the surface as early as before implantation (Charnley 1972, Lidwell et al. 1982). A third source described is the haematogenous spread of micro-organisms from distant foci in the body towards the biomaterial site. Anecdotal reports of sepsis following dental work and other bacteraemia-producing procedures like surgical incision of infectious processes are common. However, well-

documented accounts on this subject are rare (Fitzgerald and Nasser 1995, Sanderson 1991).

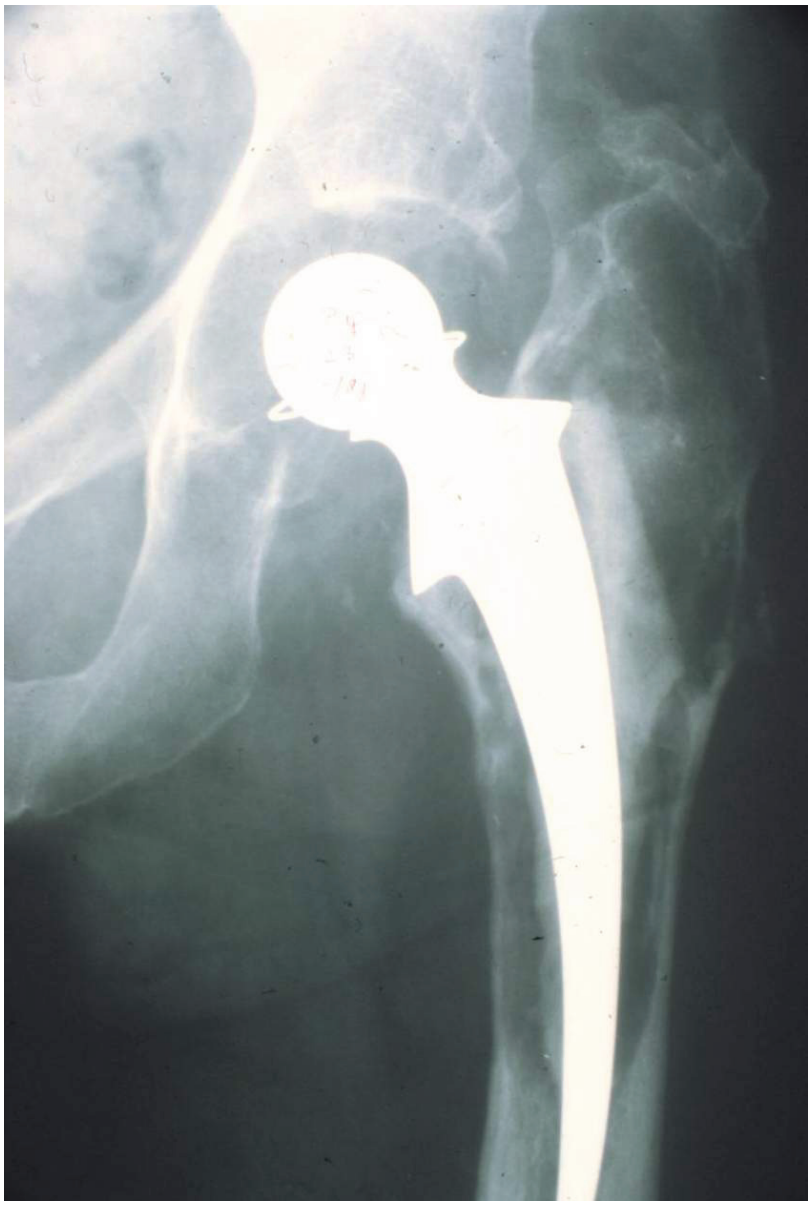

Figure 2. X-ray example of patient with a loosened cemented total hip prosthesis implanted on left side. Note the osteolysis around the femoral component.

Biomaterial-implants in permanent contact with skin and/or the outer human body environment form a class of implants that have by definition a contamination rate of 100%. This contaminated state makes them very susceptible to malfunction

because of infectious complications (Tang and Eaton 1995) (Table 1).
Clinical examples of these biomaterial-implants are intravenous catheters, peritoneal dialysis catheters, urinary tract catheters, voice prostheses, oral implants and percutaneous pins in external fracture fixation. Lower infection rates have been observed with totally implanted prostheses (Figure 2), the consequences being more serious though.

Surgical precautions and consequences

Because implants in permanent contact with skin and/or the outer human body environment have a 100% contamination rate, they have a high chance of malfunction due to infectious complications. Therefore besides the regular surgical precautions, preventive measures are being developed. This is exemplified by the coating with silver of percutaneous catheters (Davenport and Keeley 2005, Tobin and Bambauer 2003) and percutaneous pins (Masse et al. 2000). In the field of preventing infection of percutaneous pins, the use of a small electric current has proved to be effective in animal experiments (Van der Borden 2005).
The consequences of the development of a microbial biofilm can be impairment of the function of the implant or device and/or worsening of the clinical state of the patient. Because micro-organisms block the valve mechanism, a proper functioning of the voice prosthesis (Figure 3) is impaired or causes leakage of food into the trachea (Mahieu et al. 1986). An exchange procedure every 4 months of the prosthesis is the result of this process (Van den Hoogen et al. 1996). Colonization by micro-organisms of urinary tract catheters is inevitable. This can cause blockage or, more seriously, bacteriuria (Nickel et al. 1994). Infections of indwelling catheters, like for example central venous catheters, often results in bacteraemia which

can cause sepsis and endocarditis. With infections of implants in the circulatory system a high mortality rate of 50% and 70% occurs for vascular grafts and prosthetic valves respectively (Mayer and Schoenbeum 1982).

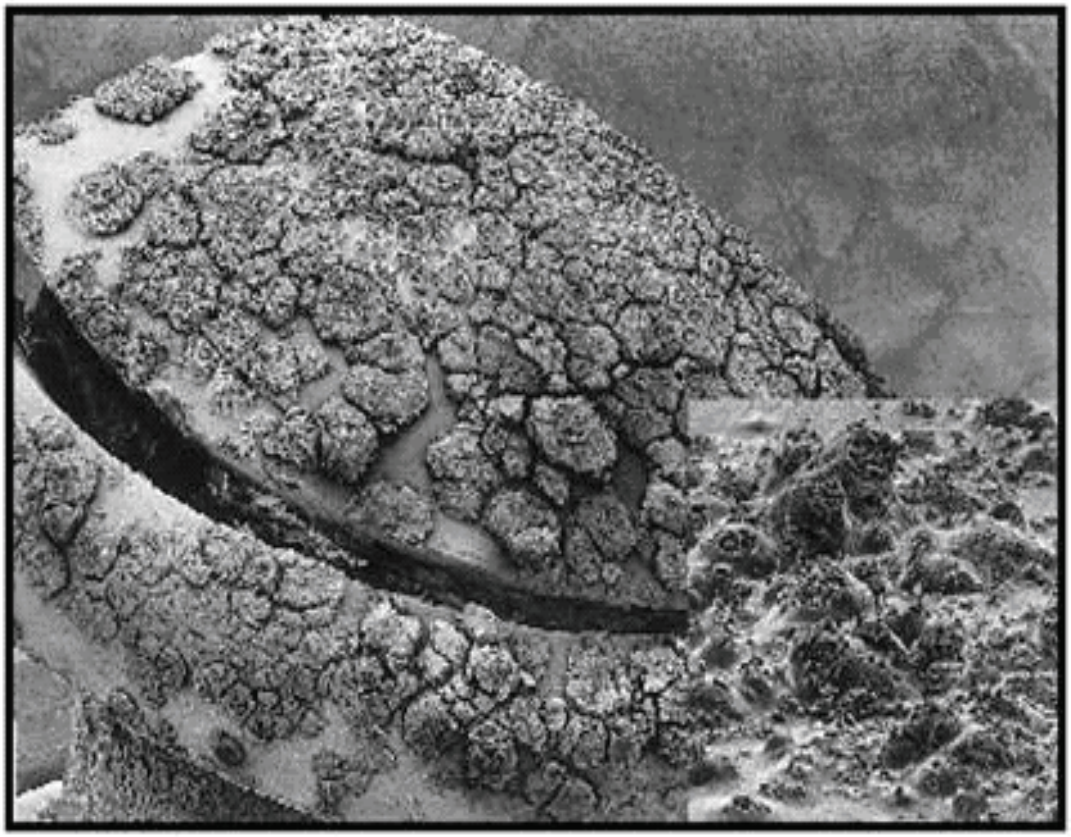

Figure 3. Example of a voice prosthesis covered with biofilm causing a malfunction in the valve mechanism.

Infection of deep tissue implants, for example orthopaedic implants, will usually result in serious complications like pain, swelling of the joint or limb and loosening of the implant, mortality rates up to 20% are reported with these kind of implants (Bengtson et al. 1987, Fitzgerald and Jones 1985, Hunter and Dandy 1977). Up to a year after microbial seeding, clinical signs of deep implant infections are being reported to appear (Maniloff et al. 1987).

This long interval between inoculation of the bacteria and the onset of symptoms can be caused by the low-virulence organisms which normally inhabit the skin and oral cavity. This may often mimic the natural "aseptic" loosening of prostheses (Costerton 2005, Phillips and Kattapuram 1983). Because of this low-virulence character of the organisms involved, in combination with the biofilm they grow in, a significant part of these infections is probably never recognized. As standard microbiological techniques are used to test the presence of infectious micro-organisms, slow growing biofilm organisms often remain undetected (Donlan 2005, Neut et al. 2003, Tunney et al. 1998 and 1999).

This has important clinical implications for the concept of "aseptic loosening" and the recurrent nature of musculoskeletal infection. Nelson et al. (2005) explained this with a sort of triple mechanism, including

(1) inadequate techniques of removing adherent, biofilm-associated bacteria;

(2) small colony forming variants; and

(3) intracellular *S. aureus* "residing" within osteoclasts.

Generally speaking a surgeon needs to perform his surgical technique well with regard to placing the incision, soft tissue handling, meticulous haemostasis and operating time, but also with regard to simple things as the application of the correct time of scrubbing hands, proper wear of hair and mouth covers and the maintenance of a strict discipline in the operating theatre. The latter aspects are most important in biomaterial-associated surgery, and because of their relative unimportance in soft tissue surgery, are frequently overlooked in implant surgery. One must realize that the most common cause of biomaterial-associated infection is thought to be peri-operative contamination (Ahlberg et al. 1978).

Avoidance of devitalisation by meticulous handling of tissue is an important variable in influencing the risks of deep infection. To prevent areas of skin necrosis between an old and a new

incision, previous incisions should be used. Local factors such as scar tissue, depending on its size and localisation, can have a decreased vascularity and it may greatly increase the time required to perform revision surgery (Charnley 1972, Klein and Cox 1994, Wilson et al. 1990). Especially when infection has been the reason for earlier operations the outcome can be adversely affected (Jerry and Rand 1988, Schmalzried et al. 1992). Meticulous haemostasis and wound closure are essential in preventing haematoma or an area of wound necrosis. Operative time has to be kept to a minimum because of the association of operative time and the development of infection (Charnley 1972).

Biomaterial-associated surgery versus non biomaterial-associated surgery

The incidence of infection after implant surgery is generally low (Table 2) and infection rates have decreased substantially over the past decades, but the often disastrous results of these infections make them important complications. Also because of the increasing incidence of for example total joint replacement infection still is a source of considerable morbidity (Okhuijsen et al. 1998).
Apart from the morbidity, the financial burden a joint prosthesis infection puts on health care systems is enormous. In the United States the annual cost to treat the 3500 to 4000 infections that develop after arthroplasties each year is between 150 and 200 million US dollars (Eftekhar 1993). In spinal surgery the use of spinal instrumentation clearly increases the risk for postoperative infection from 1% to a range of 2.1 to 8.5% (Levi et al. 1997). A large amount of the $24 billion spent in 1990 on treating spinal disorders (Schwab et al. 1995) will therefore account for the cost of treating spinal implant infections in the near future. With an increasing use of

biomaterials in surgery this financial problem will only continue to increase.

It can be argued that sterile implantation of biomaterials is virtually impossible. The operation wound is contaminated to some extent in all procedures. Minimizing contamination by optimizing the operating-room environment, protocols and the operative technique is crucial. These are the factors that can be influenced by the surgeon and the operating personnel. Performing biomaterial-associated surgery means being aware of the possibilities of contamination during the procedure. This necessitates an Operating Room (OR) discipline in operating personnel, as well as in anaesthetists, nurses, students, porters and visitors who enter the aseptic zone.

When a surgeon implants biomaterials, an important compromising factor concerning the host defence is introduced. In a classical study in man it was shown that the presence of a subcutaneous suture reduced the required inoculum to produce infection with *S. aureus* from 106 to only 200 bacteria (Elek and Conen 1957). Therewith the presence of a foreign body presents another clinical challenge on its own.

Whenever a biomaterial is introduced into the human body, surgical and mechanical trauma as well as the biomaterial itself will evoke an acute inflammatory response (Jasty et al. 1990). This acute inflammatory cascade results in localised cell necrosis and tissue degeneration and the formation of a very thin membrane between the prosthesis and the body, consisting of fibroblasts, vascular endothelium cells and macrophages. This immune response can disappear when the wound is healed and the biomaterial is encapsulated. In many cases however the host-biomaterials interface remains in a state of chronic inflammation, as few metals and plastics are completely chemically inert in the warm, wet and oxygenated environment of living tissues with a non-neutral pH, causing the release of components of the biomaterial, like corrosion products, plasticizers and monomers which are able to incite an

inflammatory reaction (Dougherty and Simmons 1982, Gristina 1987). Chronic inflammation impairs host cell growth on the implant (Jackson and Cochrane 1988) and can cause chronic pain, while it may disrupt the anchorage of the implant into the surrounding tissues thus impairing its stability leading to failing performance.

Historically orthopaedic surgeons are used to work with biomaterial implants on a large scale since the development of joint arthroplasties in the 1960's. Because they are familiar with the susceptibility of traumatized bone to infection, as has been shown in animal models of osteomyelitis (Rissing 1990, Tsukayama 1999), their OR manners and attitude towards minimizing contamination have since then been developed further and fine-tuned. Charnley already initiated this after concluding that his 7% post-operative infection rate with total hip arthroplasty was too high and operative protocols needed to be updated (Charnley 1972). Contamination of the operative wound is influenced by the OR environment. Variables affecting the OR hygienic efficiency include the number of people inside (Ritter et al. 1976) and their adherence to adequate protocols (Borer et al. 2004, Mackay et al. 2000), the amount of traffic in the OR (Ritter et al. 1976) and personnel present (Gosden et al. 1998), the preparation of the operative site (Ellenhorn et al. 2005, Seal and Paul-Cheadle 2004), the timing and technique of preoperative shaving (Klonniksen et al. 2002) and the clothing of the operating personnel (Blomgren et al. 1990, Lipp and Edwards 2002, Santos et al. 2005) including double-gloving because the chance of perforation (Tanner and Parkinson 2002) and contamination (Davis et al. 1999). Although there seems to be consensus on the importance of a clean air environment in the OR the role of laminar airflow in decreasing infection has remained controversial (Fitzgerald 1992, Lidwell et al. 1982). Some report an improvement in direct infection control (Charnley 1972, Drabu and Miller 1998, Friberg et al. 1996, Salvati et al. 1982) or indirect control by

diminishing the prevalence of contamination of the surgical instruments (Ritter et al. 1976). Others report the influence of airflow on infection rates to be less important (Espehaug et al. 1997) or to be proven (Smyth et al. 2005).
Although the above mentioned potential measures are important, the single most important variable influencing the development of postoperative implant infection is the appropriate use of peri-operative antibiotics (Antti-Poika et al. 1990, Doyon et al. 1987, Espehaug et al. 1997, Hughes et al. 1982). Peri-operative antibiotics in implant surgery are now common practice (Dent et al. 1997, Young and Lawner 1987). The type of preferred antibiotic and its appropriate regimen has been studied by Tang et al. (2003).
In addition, recording of the number of infections with feedback to the treating physician (Wong 1999) should be integrated into a registration of complications in the department, as a part of a continuous education program. This recording should preferably extend also to personnel in operating rooms, bacteriological and sterilization departments (Walenkamp 2003).

Biomaterial-associated surgery protocol

Reducing biomaterial-associated infections in surgery involves a change in the operating attitude of everyone involved in all processes that are ongoing in the OR towards decreasing contamination risks. The non biomaterial-associated surgeon is used to a more forgiving environment and therapy resistant infections are rare. Biomaterial-associated surgery by surgeons not familiar with the contamination risks and the ways of preventing them can be hazardous. To minimize these complications, the awareness of these contamination risks should be reflected in an appropriate protocol, adjusting of the peri-operative protocols and attitude of the surgeon and operating personnel.

The exact content of such a protocol is hard to ascertain, because many statements are open for debate. Looking at the essentials, however, the main goal is decreasing contamination by minimization of air disturbance. Principles for achieving this goal in a biomaterial-associated surgery protocol are: minimizing of personnel traffic in- and out the OR, personnel movement in the OR and of personnel communication.
Strict obedience by all those involved and continuous education through performance feedback together with an appropriate antibiotic prophylaxis regime should minimize the inevitable post-operative infectious complications with their devastating effect on the function and lifetime of the biomaterials involved as well as on the patient who is the victim.

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3

PER-OPERATIVE CONTAMINATION IN PRIMARY TOTAL HIP ARTHROPLASTY

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Abstract

All surgical procedures have the risk of microbial contamination. However, procedures in which prosthetic materials are involved carry a high risk for future infectious problems because of the protection offered by the biofilm mode of growth. Per-operative contamination studies have been conducted on involved surgical instruments, but whether these instruments transmit the contamination to the prosthesis or future site of the prosthesis can only be guessed. The aim of this study was to detect possible bacterial contamination in total hip arthroplasty through instruments that are used at the direct site of implantation during the primary procedure. Samples of the broaches used for preparing the acetabulum and femur, as well as samples of the reamed acetabular and femoral bone, were collected during 67 consecutive primary total hip arthroplasties in 67 patients. Broach samples were taken at the start and end of every reaming procedure. The total number of samples taken amounted to 402, of which 26 were found to be positive for micro-organisms. In 20 patients at least one of these positive samples had been in direct contact with the actual prosthesis site, indicating that at least 30% of the involved patients had a possible bacterial contamination when leaving the operating theatre.

Introduction

Microbial contamination is a potential hazard of all surgical procedures, which can result in septic complications. Procedures in which biomaterials are involved especially have a high risk for future infectious problems because of the protection offered by the biofilm mode of growth of the organisms on the prosthesis. The host defence is considerably compromised in the presence of a foreign material (Elek and

Conen 1957, Lidwell et al. 1983[b], Zimmerli et al. 1982), and otherwise relatively low-virulent organisms, such as *Staphylococcus epidermidis*, become a common infecting organism in biomaterial-associated infection (Christensen et al. 1989). Eventually, septic complications can force the surgeon to remove the implant.

Per-operative contamination has for many years been considered the most common cause of biomaterial-associated infection (Ahlberg et al. 1978, Glynn and Sheehan 1983, Lidwell et al. 1983[a]). The operation wound and implant surface can be readily reached by micro-organisms through diffusion, active movement, or haematogenous transport (Baird et al. 1984, Ha'eri and Wiley 1980, Lidwell et al 1983). In addition, instruments can become contaminated by bacterial deposition from air or by skin contact and subsequently enter the wound through contaminated instruments (Christensen et al. 1989, McCue et al. 1981, Strange-Vognsen and Klareskov 1988). Strategies have been tried to diminish this by washing or storing instruments in a splash basin. This strategy unfortunately had an adverse effect (Baird et al. 1984). Consequently, it can be concluded that, despite the use of pre-operative systemic antibiotic prophylactics, strict hygienic protocols, sterile operating theatres, and special sterile enclosure, prostheses inevitably become contaminated during surgery. Per-operative contamination studies have been conducted on involved surgical instruments, such as gloves (28.7%) (Davis et al. 1999, Sanders et al. 1990), light handles (14.5%) (Davis et al. 1999, Robinson et al. 1993), knives (9.4%) (Davis et al. 1999), and suction tips (11.4%) (Christensen et al. 1989, Greenough 1986). However, only gloves and to a lesser percentage also suction tips come in close contact with the actual prosthesis site or prosthesis itself. No studies have been performed to detect possible bacterial contamination of instruments that have had contact with the actual implant site such as broaches for preparing the

acetabular and femoral bone. Bacterial contamination of these instruments may likely be considered as a baseline for contamination in the operative field and micro-organisms involved.
Therefore, the aim of this study was to detect possible per-operative bacterial contamination in Total Hip Arthroplasty (THA) through instruments used at the direct site of the prosthesis during the primary procedure.

Material and Methods

In the period from July 2001 until December 2002 during 67 consecutive primary THAs in 67 patients, samples of the broaches used for preparing the acetabulum and femur were collected, as well as samples of the reamed acetabular and femoral bone. Standard preoperative care consisted of painting the skin twice with iodine tincture (iodine 1% in alcohol 70%; Fresenius Kabi, 's-Hertogenbosch, The Netherlands) without scrubbing and antibiotic prophylaxis with 1000 mg Kefzol, cefazolin (Eli Lily Nederland BV, Houten, The Netherlands) given intravenously at the time of the induction of anaesthesia. All operations were performed under vertical laminar flow, impervious non-iodine impregnated skin drapes and the operating team wore disposable impervious drapes. A total of six samples were collected during every operative procedure (Table 1). Splash basins were not used during the procedure. At the time of the incision the smallest unused acetabular broach was sampled (Sample 1). After sampling, the acetabular reaming procedure was started with this particular broach. At the end of the acetabular reaming, a sample from the largest unused acetabular broach was taken (Sample 2). This broach was never used at the direct site of the prosthesis.

Table 1. Different broach samples and bone chips used for culturing in 67 patients requiring an orthopaedic implant and the number of positive samples.

Sample number	Description	Number of positive cultures	Contact with prosthesis site
1	Smallest unused acetabular broach	6	Yes
2	Largest unused acetabular broach	2	No
3	Smallest unused femoral broach	2	Yes
4	Largest unused femoral broach	4	No
5	Removed acetabular bone	5	Yes
6	Removed femoral bone	7	Yes
Total positive samples		**26**	

Before starting the reaming procedure of the femur, a sample was taken from the smallest unused femoral broach (Sample 3). After sampling, the femoral reaming procedure was started with this particular broach, and at the end of the femoral reaming, a sample was taken from the largest unused femoral broach (Sample 4). This broach was never used at the direct site of the prosthesis. All broach samples were taken with sterile swabs (COPAN, Italy). These swabs are delivered with a sterile tube containing an agar gel suitable for sterile transport of aerobic and anaerobic micro-organisms. Before sampling, the swabs were moisturized in a sterile 0.9% NaCl solution, and kept in a closed container. Acetabular (Sample 5) and femoral (Sample 6) bone removed during reaming were collected in a culturing medium and sent in for microbial evaluation. Trypton soya broth (TSB, Oxoid, Basingstoke, UK) was used for

collecting and culturing the bone removed by reaming. All samples were transported to the laboratory within 24 hours and the material swabbed from the broaches was cultured on enriched blood agar (BA) plates (+0.5% hemin and 0.1% menadione). All material was incubated at 37°C both aerobically and anaerobically for 7 days. When growth was present on either the BA plates or in the TSB containers, samples were taken for Gram-staining. Gram-positive cocci were subjected to a catalase and DNase test to identify coagulase-negative staphylococci (CNS) and *Staphylococcus aureus*.
During the study, three sham culturing procedures during a comparable operation were performed at different times. In 3 planned aseptic revisions of a THA the same culturing protocol as previously described was used. The sham culturing procedures differed from the original protocol in only two ways. First, after moisturizing swabs in the closed container containing a sterile 0.9% NaCl solution, the swabs were put in their sterile tubes and were sent in without sampling the broaches. Secondly, the TSB containers were opened and closed again without leaving retrieved material behind.

Results

The total number of direct samples taken in the field of operation amounted to 402. Of these samples only the ones numbered 1, 3, 5 and 6 where considered a possible source of direct contamination. Table 1 shows the samples taken that were found positive. 26 samples were contaminated with viable micro-organisms (6.5%). These positive samples were found in 21 patients. In 20 patients at least one of these positive samples had been in direct contact with the actual prosthesis site (Sample Numbers 1, 3, 5 and 6); indicating that 20 of the 67 patients involved (30%) had acquired contamination with a micro-organism of their future prosthesis site. No micro-

organisms were cultured from any of the sham procedures, indicating no contamination during transport and handling in the laboratory. There was no clear evidence that samples taken in a later time during procedure showed a higher rate of contamination than the ones that were taken at the beginning of the procedure.

A total of 26 samples contained 28 different microbial strains (Table 2), while by consequence 2 samples showed 2 different micro-organisms. Coagulase-negative staphylococci (CNS) were cultured 13 times, whereas *S. aureus* was determined twice. Rods were cultured 10 times, of which seven were Gram-positive micro-organisms. Those micro-organisms that could not be classified as CNS, *S. aureus* or rods were left unidentified. This group consisted of two Gram-positive coccal strains and one fungal strain. The fungal strain was identified microscopically without specific culturing techniques.

Table 2. The strains and species isolated from broach samples and bone chips used for culturing in 67 patients requiring an orthopaedic implant

Micro-organism	**Number**
Coagulase-negative staphylococci	13
Gram-positive rods	7
Gram-negative rods	3
Staphylococcus aureus	2
Gram-positive cocci	2
Fungus	1
Total	28

Discussion

The most common cause of biomaterial-associated infection in orthopaedics is thought to be per-operative contamination (Ahlberg et al. 1978, Glynn and Sheehan 1983, Lidwell et al. 1983[a]). The operation wound and implant surface can be reached by micro-organisms through several ways. In the past, several per-operative contamination studies have been performed, all showing a variable percentage of contamination of several instruments (Christensen et al. 1989, Davis et al. 1999, Greenough 1986, Sanders et al. 1990). A disadvantage in these studies is the fact that it is only postulated that contamination of these instruments will cause contamination of the prosthesis, while none of the instruments is actually in direct and close contact with the implantation site. This yields of course the potential of contamination, but it would be preferable to take cultures from the actual implantation site as well, as done in this study. This study investigated the possible contamination risk of primary THA directly through instruments used at the planned site of the prosthesis, including bone removed from the implantation site.

Viable micro-organisms were cultured from instruments and material that had been in direct contact with the actual site of the prosthesis in 30% of patients involved in this study, which is comparable with 28.7% glove contamination in the recent study of Davis et al. (1999). This suggests that there is a close relationship between glove and implant site contamination.

Possible bacterial contamination of the implant site is a direct threat to the implanted prosthesis. Gristina et al. (1987 and 1988-1989) described the faith, i.e. success or failure, of a biomaterials implant in the human body as a "race for the surface" in which tissue integration competes with microbial adhesion. Consequently, 30% of the patients involved in this current study must be considered at risk when leaving the

operating theatre, because infecting micro-organisms were introduced before tissue integration could commence.
In the study of Davis et al. (1999) as well as in the present study, CNS was found to be the main contaminating organism in 76% and 46% (13 out of 28) of the cases, for the study of Davis et al. (1999) and ours, respectively. It is interesting to dwell on the question whether this bacterial contamination of the implant site during surgery has clinical consequences. One problem in answering this question is that up to 1 year after microbial seeding, clinical signs of deep implant infections are being reported to appear (Maniloff et al. 1987). This long interval between inoculation of the bacteria and the onset of symptoms may have a variety of different causes that are difficult to distinguish from each other, of which low-virulence organisms introduced during insertion of the prosthesis is one. In this respect, also aseptic loosening of prostheses (Neut et al. 2003) might be due to adhesion of low-virulence organisms. Likely, a considerable part of these infections is never recognized.
In a recent study, primary arthroplasties of the hip and knee were found contaminated with bacteria during surgery, but no reflection of the contaminating strains was found at the time of revision (Davis et al. 1999). This could well be due to inadequate culturing methods impeding detection of low-grade infections by slowly growing biofilm organisms (Neut et al. 2003, Phillips and Kattapuram 1983, Tunney et al. 1998 and 1999). In this study, follow-up of the patient population revealed that in the non contaminated group, patients had no infectious complications within nearly two years after surgery. 2 patients, contaminated per-operatively, had persisting complaints that, together with laboratory and skeletal scintigraphy findings, suggested a low grade infection. In both patients, *S. aureus* was found contaminating the broaches during surgery.

Summarizing, in the current study, 30% of the primary THAs were placed in a site contaminated with bacteria before the insertion of the prosthesis. We feel that this study provides a good baseline for bacterial contamination of the actual implantation site and that this early contamination might be indicative for the occurrence of prosthetic infection. Splash-basins filled with a salt solution have hitherto not worked well to reduce instrument contamination, but possibly splash-basins filled with an antimicrobial solution like chlorhexidine might help to reduce the problem of per-operative bacterial contamination.

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4

DETECTION OF BIOMATERIAL-ASSOCIATED INFECTION IN PRESUMED ASEPTIC LOOSENING OF JOINT PROSTHESES

Abstract

We have retrieved 33 prostheses or prosthetic components from patients requiring revision surgery due to presumed aseptic loosening. The components involved were transported in sterile bags to the laboratory for an extensive microbiological culturing of the prosthetic surfaces (aerobic and anaerobic culturing of scrapings for 7 days). Simultaneously, tissue samples were excised for extensive culturing. In only one of the 33 cases, micro-organisms were detected by routine culturing of tissue, while extensive culturing demonstrated infectious organisms in tissue samples from 14 cases. In addition, extensive culturing of the biomaterials scrapings identified 6 other cases of positive cultures, totalling the percentage of infected cases by extensive culturing to 60% (20 out of 33 patients). These results demonstrate that biomaterial-associated infections may well remain undetected by standard clinical and microbiological hospital procedures.

Introduction

The number of patients requiring an artificial joint has grown rapidly to more than 1.3 million people in the United States (Praemer et al. 1992). Approximately 20% of all joint replacements fail (Christel and Djian 1994), yielding prosthesis removal and revision with concomitant patient trauma and increased medical costs (Dreghorn and Hamblen 1989). Aseptic loosening is the most common cause of prosthetic joint failure reported (Malchau et al. 2004). Standard orthopaedic practice is to exchange aseptically loosened joint prosthetic components in a one-stage procedure. Unfortunately, after revision, the rate of infection is higher than after primary procedures, probably due to presence of unrecognised infection at the revision (Dupont 1986). As shown by different studies, biomaterial-

associated infections differ from other infections because of the presence of a biofilm (Gristina 1987, Gristina et al. 1988, Neut et al. 2003). Up to a year after microbial contamination of an implant by biofilm organisms, clinical signs of deep implant infections are being reported to appear (Maniloff et al. 1987). This long interval between first contamination and the onset of symptoms, may have a variety of different reasons, of which low-virulence organisms introduced during insertion of the prosthesis constitute one. In this respect, also aseptic loosening of prostheses might be due to adhesion of low-virulent organisms, such as from the skin and dental microbiota. Likely, a considerable part of these infections is never recognized (Tunney et al. 1998 and 1999, Nelson et al. 2005), yielding the diagnosis "aseptic" loosening (Phillips and Kattapuram 1983). Neut et al. (2003) showed recently that the detection rate of infectious organisms in septic-loosening can be improved by extensive culturing of scrapings from the implant surfaces (briefly described as a biomaterial based extensive culturing procedure).

The aim of the current study was to investigate whether this extended biomaterial-based culturing procedure, when applied in situations with the presumed diagnosis "aseptic loosening", could justify a change in attitude and culturing protocols in revision situations where doubt arises on the aseptic cause of the loosening.

Patients and Methods

Patient group and clinical procedure

33 prostheses or prosthetic components after presumed aseptic loosening were retrieved from patients during revision surgery in the period June 1999 to May 2001. This study was approved by the Institutional Review Board for human experiments.

Table 1. Bacterial strains isolated from the prosthetic components during aseptic revision.

Micro-organism	**Number of times cultured**
Coagulase-negative staphylococci (CNS)	14
Gram-positive rods	2
Gram-negative rods	3
Staphylococcus aureus	4
Other Gram-positive cocci	11
Total	34

Patients included 27 women and 6 men with a mean age of 68 years (27 to 93) and a mean interval from previous joint placement to revision surgery of 11.3 years (1.3 to 17.5 years). In the study population, the revised components were 12 Total Hip Arthoplasties (THA), 3 femoral components of a THA, 9 acetabular components of a THA, 6 Total Knee Arthroplasties (TKA), 2 tibial components of a TKA and one Unilateral Knee Arthroplasty (UKA). In 5 cases, patients already had prior revision surgery. All patients had radiological and clinical signs of loosening of their joint prosthesis. The mean pre-operative value of ESR was 15 mm/h and the mean pre-operative CRP level was 7 mg/l.

Standard preoperative care consisted of painting the skin with Betadine and antibiotic prophylaxis of 1500 mg Cefuroxim EB (Eurobase B.V., Barneveld, The Netherlands) given intravenously at the time of the induction of anaesthesia. All operations were carried out under vertical laminar flow and the operating team wore disposable impervious drapes. The implants were removed and placed aseptically in a sterile bag, filled with Reduced Transport Fluid (RTF) (NaCl 0.9 g/l, $(NH_4)_2SO_4$ 0.9 g/l, KH_2PO_4 0.45 g/l, $MgSO_4$ 0.19 g/l, K_2HPO_4 0.45 g/l, Na_2EDTA 0.37 g/l, L-Cysteine.HCl 0.2 g/l, pH 6.8).

Tissue in contact with the implants was excised and swabbed. This material was divided for use in the routine hospital culturing and the extended culturing procedure.
Routine hospital culturing consists of culturing material in thioglycolaat medium for 3 days and on blood agar (BA) for 2 days both at 35°C in an aerobic incubator. Cultures on anaerobic blood agar (ABA) plates were grown for 4 days at 35°C in an anaerobic incubator. Implants and excised material was transported to the Department of Biomedical Engineering for extensive culturing within 2 to 24 h after removal. After transport, prosthetic surfaces were scraped with a sterile knife to dislodge biofilm bacteria. Scrapings and excised tissue were cultured on enriched BA plates (BA +0.5% hemin and 0.1% menadion), incubated at 37°C both aerobically and anaerobically for 7 days.

Microbiological determination

All organisms isolated were Gram stained. Rods were microscopically identified as Gram-positive or negative. Gram-positive cocci were subjected to a catalase (hydrogen peroxide solution 3%) and deoxyribonuclease test (Dnase agar, Oxoid, Basingstoke, Great Britain). Strains were identified as Coagulase Negative Staphylococci (CNS) from a positive catalase and negative DNase-test. When both the catalase and the deoxyribonuclease tests were positive, isolates were identified as *Staphylococcus aureus*.

Results

Clinical microbiological culturing of the tissue samples showed only a positive culture in one patient (3%). Extensive culturing of tissue samples showed bacterial growth in 14 patients (42%), whereas extensive culturing of biomaterial scrapings

indicated bacterial growth on prosthetic components retrieved from 12 out of 33 patients (36%). In 6 of these patients bacterial growth was already demonstrated from tissue cultures, and consequently, 6 additional, infected patients (18%) were identified from cultures of implant surface scrapings. This totals the percentage of infected cases in suspected aseptic loosening by extensive culturing to 60% (20 out of 33 patients).
An overview of micro-organisms cultured is given in Table 1. CNS is the most frequently encountered micro-organism, being cultured in 14 out of 34 positive samples. However, also other Gram-positive cocci were frequently found in 11 samples.
S. aureus was only found in 4 cases.
From 10 components only 1 microbial strain was cultured. From 7 components 2 different contaminating species were found, while from 4 components 3 different species were cultured.

Discussion

In the current study, joint replacement components removed because of aseptic loosening were examined for the presence of infectious bacteria. Clinical microbiological culturing of excised tissue showed only 1 positive culture in 33 patients (3%). Extensive culturing tissue samples yielded 42% positive cultures and extensive culturing of biomaterials scrapings added another 18% totalling to 60%, with CNS being the predominant causative organism.
In the workup for revision surgery, extensive effort is made in distinguishing between septic or aseptic loosening, because the consequences for subsequent therapy are different. However, differentiation between septically or aseptically loosened joint prostheses is often difficult. One problem in this differentiation is that up to 1 year after microbial seeding, clinical signs of deep implant infections are being reported to appear (Maniloff

et al. 1987) and in this respect, also septic loosening (Neut et al. 2003) might be due to adhesion of low-virulence organisms. It seems likely therefore that a considerable part of these infections is never recognized (Nelson et al. 2005).
This puts any one stage aseptic revision at risk. During revision surgery, a choice has to be made between lengthening the procedure to extract hard to reach debris or shortening the procedure to leave these presumed "aseptic" parts in situ. This study shows that the extra effort for removing "aseptic" debris could be worthwhile in preventing the ongoing process of infection in 60% of the cases. If during the revision surgery, however, doubt arises concerning the aseptic character of the loosening, the surgeon should always change from the one stage to a two stage setup. Material removed should then be cultured according a biomaterial based extensive culturing protocol as this yields more identification of infecting organisms than routine hospital culturing of tissue. Atkins and Bowler (1998) found that by extending the culturing time to 7 days, the detection rate of infectious bacteria in excised tissue samples could be increased to 64%.
Several methods, varying from immunofluoresence microscopy, PCR amplification and ultrasonication (Tunney et al. 1988 and 1999), have been used to improve the detection rates in orthopaedic implant loosening. Although being highly sensitive, these methods are difficult to introduce to standard clinical practice because of their complexity and high financial burden. Moreover, good adequate microbiological culturing should in principle be sufficient (Ince et al. 2004).
One important remark has to be made though, about the per-operative circumstances under which these cultures are taken. Recent research in our hospital showed that in primary THA the per-operative circumstances resulted in a 30% contamination of the direct site of the prosthesis (Maathuis et al. 2005). In a subsequent study from the same institution a comparable contamination percentage was found (Knobben et al. 2006).

Per-operative contamination may therefore not a priori be ruled out in revision surgery either and a true aseptically loosened prosthesis may become contaminated during removal with an impact on the present results.
We improved our detection rate from 3 to 60% by scraping the surface of the implant and culturing tissue from the peri-prosthetic area for a longer period. In normal clinical practice, the standard hospital cultures would have confirmed our diagnosis of aseptic failure of the biomaterial and no further supplemental treatment would have been proposed. The method presented in this chapter is easy to use in every hospital situation where prosthetic revision procedures are being performed and does not add extra costs to the budget of the hospital involved. On the basis of the current results, hospital diagnoses in orthopaedic revision surgery should be made by analysis of scrapings of the biomaterial surface of prostheses, which will increase the detection rate of intra-operatively suspected prosthetic joint infections. Any organisms colonizing the primary prosthesis, but not identified and eradicated, will likely infect a new implant and put the revision surgery at risk of failure. Follow up of this study population will be needed to detect the clinical relevance of these findings.

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5

SEPTIC OR ASEPTIC LOOSENING OF JOINT PROSTHESES
A pilot study with 18FDG-Positron Emission Tomography

Introduction

Pain is an important symptom of loosening of a joint prosthesis. Loosening of joint prostheses may be septic or aseptic, and the consequences for subsequent therapy are different. However, differentiation between septic or aseptic loosened joint prostheses is often a difficult clinical problem. Several diagnostic techniques are being used nowadays to detect or exclude infection. Plain-film anteroposterior and lateral radiographs are the most common form of imaging. Despite their widespread use, these radiographs are rarely diagnostic in the differentiation between septic or aseptic loosening (Weisman 1983). Laboratory evaluation will usually show an elevation of the erythrocyte sedimentation rate (ESR) and of the C-reactive protein (CRP) (Sanzén and Carlsson 1989) in the case of an infection, but in general such studies are suggestive, not diagnostic. Scintigraphy has gradually become a more reliable adjunct in evaluation of a painful joint prosthesis. ^{99}Technetium–diphosphonate bone scintigraphy, ^{67}Gallium or ^{99}Technetium-labeled leukocyte scintigraphy are helpful options. None of these techniques is sufficiently sensitive or specific in diagnosing infection (Levitsky et al. 1991). Consequently, combinations of these techniques are often being used to improve the diagnostic accuracy (Palestro et al. 1990). A more invasive procedure to discriminate between pain originating from a septically or aseptically loosened joint prosthesis is aspiration of artificial joint fluid. This technique has been judged as controversial because of the lack of sufficient sensitivity (Hanssen and Rand 1999), varying between 60% (Barrack and Harris 1993) and 100% (Duff et al. 1996) depending on the method used. Insufficient sensitivity may be related to the fact that infectious organisms in biomaterial associated infections predominantly grow in a "biofilm" mode (Chimento et al. 1996). After implantation of

biomaterial in the human body a "race for the surface" starts (Gristina 1987, Gristina et al. 1988) in which tissue-integration competes with microbial adhesion. When micro-organisms win this race and adhere to the biomaterials surface subsequent surface growth of the micro-organisms will lead to a mature biofilm and infection. This makes it difficult to aspirate the causative organism by puncture of the involved artificial joint since this is not where they predominantly reside. Besides this, invasive procedures carry the risk of introducing pathogenic organisms into a previously aseptic joint.

Positron emission tomography (PET) with the radio-pharmaceutical 2-[^{18}F]fluoro-2-deoxy-D-glucose (FDG) is nowadays widely used in oncology. Its use is based on an up-regulation of glucose-transporters (in particular GLUT-1) on the cell membrane of the oncocyte, which causes an increased uptake of glucose in the cell (Chung et al. 1999). FDG behaves as glucose, however, and after its phosphorylation, it is trapped within the cell, thus causing an entrapment of FDG within the cell, especially the oncocyte. FDG has been shown to be a very sensitive indicator of the presence of malignancy. In contrast, specificity, although also being high, is still far from perfect. It has been shown that FDG also accumulates in inflammatory tissues, particularly in macrophages (Kubota et al. 1992). Recently, this has led to a concept of diagnosing inflammation with FDG-PET, e.g. in fever of unknown origin (Lorenzen et al. 2001). FDG-PET has also been addressed in recent literature as a helpful adjunct in diagnosing musculoskeletal infections (Winter et al. 2001, Zhuang et al. 2001).

The purpose of this pilot study was to evaluate the feasibility of using FDG-PET for the differentiation between septic and aseptic loosening of joint prostheses.

Materials and Methods

In this prospective pilot study, 7 patients with a painful joint prosthesis (2 Total Knee Arthroplasties (TKA), 3 hybrid and 2 uncemented Total Hip Arthroplasties (THA)) constituted the study group (see Table 1). All prostheses were at least one and a half years in situ (1.5-7.5 years). The study was approved by our institutional review board. Patients were older than 18 years, had no diabetes and gave informed consent. The PET study was conducted with an ECAT951/31 camera (Siemens/C.T.I., Knoxville, U.S.A.). This camera has a patient aperture of 56 cm in diameter and acquires 31 planes over a 10.8 cm field-of-view. On the day of the investigation, the patient had to refrain from food for at least 6 h before the scan. Drinking of non-calorie beverages and continuation of medication was permitted. An attenuation corrected PET study in Whole Body mode was made over the area of interest, 90 min after injection of 400 MBq FDG via an intravenous canula. No muscle relaxants were applied, but patients were instructed to stay seated during the waiting period in order to reduce physiologic muscle uptake. All patients underwent a revision procedure of their joint prosthesis within 6 weeks after this investigation.

In addition, a group of 5 patients with 7 asymptomatic joint prostheses (4 cemented, 1 uncemented THA and 2 Hemiprostheses) that went through a PET scan for oncological problems served as a control group (see Table 2). The procedure of scanning was similar as in the group with a painful joint prosthesis; however, the field of view was extended in order to cover the body from the head to the mid-thigh.

Data analysis

Radiographs were studied for radiographic signs of instability. For this purpose the Knee Society Roentgenographic Evaluation System (Ewald 1989) was used for evaluating the radiographs of the patients with a TKA. For the cemented stems of the THA the scoring system according to Gruen was used (Gruen et al. 1979). For the cemented sockets, the DeLee-Charnley classification (DeLee and Charnley 1976) was used. For the stems in the uncemented THA the criteria according to Vresilovic (Vresilovic et al. 1994) were used to ascertain stability. Because literature does not provide a separate scoring system for uncemented cups, the principles of Vresilovic (Vresilovic et al. 1994) and Engh (Engh et al. 1990) were used to interpret the radiographic signs on these radiographs.

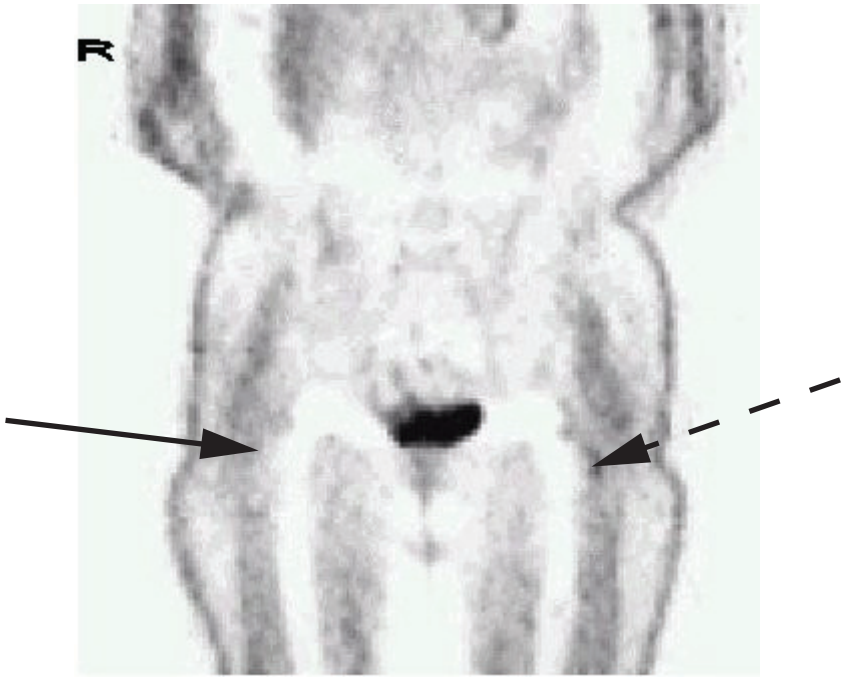

Figure 1. Example of grade 0 FDG activity around hemiprosthesis on the right side (——►). On the left side is a hemiprosthesis with grade 1 FDG activity around the greater trochanter region (- - - -►).

PET images were scored on a 3-point scale based on the intensity of the FDG uptake of the images: 0 (no uptake, see Figure 1), 1 (mild uptake, see Figure 2) and 2 (intense uptake, see Figure 2). Two experienced, independent specialists in Nuclear Medicine evaluated the images. Both readers were aware whether the scan was made because of problems with the joint prosthesis or for oncological reasons. The readers were blinded for further clinical symptoms of the patients. In case of the control group, they were aware the patients had no reported problems with their joint prostheses. In case of disagreement between readers a final verdict was reached by consulting a third reader.

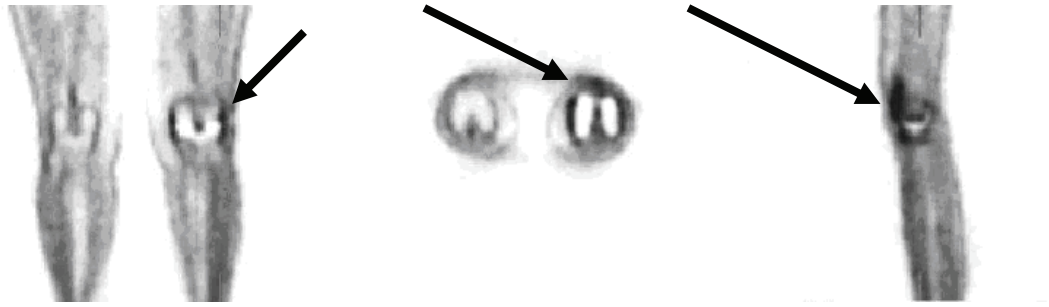

Figure 2. Example of grade 2 FDG activity around a left total knee prosthesis (⟶).

In the study group, the final diagnosis was made by surgical exploration. Routine hospital culturing was performed, meaning aerobic and anaerobic culturing for a period of 4 days. PET results were compared with the microbiological results in those patients of the study group (See Table 1 and 2). Obviously, no microbiological material was obtained from the patients of the control group, since these patients had no clinical signs of loosening of their joint prostheses at the time of scanning and none had radiographic signs of loosening.

Results

Study group: Patients with symptomatic joint prostheses

Of the 7 patients with a painful joint prosthesis 4 had category 2 FDG uptake. After revision, 3 were diagnosed as septically loosened. In one patient, both *Staphylococus aureus* and Coagulase Negative Staphylococci (CNS) were cultured. The other two patients had both CNS cultured from the tissue acquired during revision.

Table 1. Overview of patient population and data in study group.

Study group					
Type of prosthesis	Time to scan	Prosthesis painful	Laboratory findings	Activity grade on PET scan	Culture result
TKA	2 y	Yes	ESR=12 CRP=5	2	CNS
TKA	2 y	Yes	ESR=10 CRP=17	2	CNS
Uncem. THA	6 y	Yes	ESR=49 CRP=24	2	*S. aureus* CNS
Proplast® THA	12 y	Yes	ESR=12 CRP=3	2	NEG
Hybrid	5 y	Yes	N.A.	1	NEG
Hybrid	8 y	Yes	N.A.	1	NEG
Uncem. Cup	14 y	Yes	CRP < 4	1	NEG

Hybrid=uncemented cup with cemented stem, THA=Total Hip Arthroplasty, TKA=Total Knee Arthroplasty, Uncem=uncemented, NA=not available, ESR=erytrocyte sedimentation rate(in mm/h), CRP=C-reactive protein(in mg/l), CNS=Coagulase Negative Staphylococci, NEG=negative, 0=no uptake, 1=slightly increased uptake, 2=intensely increased uptake, POS=positive

The one patient with category 2 FDG uptake, but no micro-organisms cultured, had a pseudo-infectious-like mass around his uncemented femoral stem (a Proplast® femoral stem).
Three patients with a painful joint prosthesis had category 1 FDG uptake on the PET scan. Standard hospital cultures of tissue acquired during revision were not able to detect micro-organisms and these 3 prostheses were diagnosed as aseptically loosened (see Table 1).

Table 2. Overview of patient population and data in control group.

Control group					
Type of prosthesis	Time to revision	Prosthesis painful	Laboratory findings	Activity grade on PET scan	Culture result
Hemiprosthesis	5 y	No	ESR=6	0	#
Hemiprosthesis	4 y	No	ESR=6	1	#
THA	16 y	No	ESR=47	1	#
THA	16 y	No	ESR =47	1	#
THA	1 y	No	ESR =13	1	#
THA	Lost to follow-up	No	N.A.	1	#
Uncem THA, Mallory Head®	5 y	No	ESR=28 CRP=38	2	#

THA=Total Hip Arthroplasty, TKA=Total Knee Arthroplasty, Uncem=uncemented, NA=not available, ESR=erytrocyte sedimentation rate(in mm/h), CRP=C-reactive protein(in mg/l), CNS=Coagulase Negative Staphylococci, NEG=negative, 0=no uptake, 1=slightly increased uptake, 2=intensely increased uptake, POS=positive, # =no culture obtained

Control group: Patients with asymptomatic joint prostheses

The FDG uptake around the 7 asymptomatic joint prostheses varied between category 0 (N=1 hemiprosthesis), category 1 (N=5, of which 1 hemiprosthesis and 4 THA) and category 2 (N=1 uncemented THA, Mallory Head®). As these subjects were scanned for other reasons than their joint prosthesis, no microbiological confirmation was obtained (see Table 2).

Discussion

In this study we examined the feasibility of differentiating aseptic and septic loosening of joint prostheses with FDG-PET. In order to do so, it is needed to have an idea on the uptake of FDG around asymptomatic prostheses. Our data showed a mild uptake in the majority of patients, and even intense uptake in 1 asymptomatic patient, who had received an uncemented THA, Mallory Head®), placed 5 years before PET-scanning. This suggests the continuation of a reaction or inflammation after introduction of a joint prosthesis.

Theoretically, whenever a joint prosthesis is introduced into the human body, surgical and mechanical trauma will evoke an acute inflammatory response. This acute inflammatory cascade results in localised cell necrosis and tissue degeneration. Because of these processes, a so-called interface, i.e. a very thin membrane between the prosthesis and the body, consisting of fibroblasts, vascular endothelium cells and macrophages, is formed. Since it is known that macrophages accumulate FDG, it is reasonable to assume that activity can be seen on PET images even when no infection is present (Kubota et al. 1992). Consequently, the interface must not be considered as inert but as an activated tissue, with its own

ability to show activity on metabolic activity studies such as FDG-PET. Therewith, the question can be raised whether FDG-PET is able to differentiate between aseptic and septic loosening (Winter et al. 2001), since a background of slightly increased FDG uptake is imaginable (Zhuang et al. 2002). However, if infection is involved in the loosening process, a summation of activities takes place, resulting in a more intense reaction that allows differentiation between aseptic and septic loosening.

One could argue that one of the prosthesis with grade 2 activity had no proof of a septic loosening. During revision though, this procedure was converted from a one stage into a two-stage procedure. This conversion was chosen because of the intraoperative findings of signs of an infectious-like mass. Further analysis showed that this Proplast® stem had inflicted a very intense granulomatous reaction, mimicking an infectious process. This granulomatous reaction in this type of prosthesis has been described before (Maathuis and Visser 1996). This intense reaction may have caused a possible summation of FDG activity in PET scanning.

In line with this remark is the fact that an uncemented joint prosthesis inflicts a more intense reaction than a cemented one. Because of bone ingrowth, uncemented prostheses will cause, more pronounced osteoblastic activity in comparison with cemented ones (see Table 1 and 2), as has also been demonstrated using ^{99}Technetium bone scans of these two types of joint prostheses (Maniar et al. 1997).

Microbiological confirmation of infection was lacking in three patients with painful arthroplasties, of which one had a score 2 FDG activity, which might be due to a low-grade infection of the joint prosthesis that remained undetected by routine hospital microbiological techniques, as common in our hospital during the time of this study. Indeed low-virulence and low metabolically active organisms are often never recognized with standard microbiological techniques (Tunney et al. 1998, 1999, Neut et al. 2003) and estimates are that 15 to 20% of all a-

septically diagnosed loosenings are in fact septic (Mariani et al. 1996, Tunney et al. 1998). In our hospital we have improved our detection rate in periprosthetic infections by culturing peri-prosthetic tissue and biofilm from the prosthesis surface for 7 instead of 4 days (Neut et al. 2003). Unfortunately in the current study this extensive culturing method was not in use, as it might have improved the correspondence between microbiological confirmation, painful arthroplasties and FDG activity monitored. Loose joint prostheses, whether infected or not, inflict a reaction in the adjacent tissues. This reaction is visualised by a positive reading on the PET images. An intense grade 2 reaction is suggestive of a possible summation of different kind of processes. Around a joint prosthesis that causes pain to the patient this can be suggestive of an infectious process.

At this stage it is too early to make statements on the cost-effectiveness of including an FDG-PET into the work-up of patients with problems of joint prosthesis. Statements can only be speculative at this stage. Theoretically, there is the opportunity of altering the approach of the treatment in case of severely infected prosthesis. However, to do so not only a high sensitivity but also a high specificity is required. Although our results, and those of others, are promising in this respect, no final verdict can be given yet and further research into the matter is needed before FDG-PET can routinely be introduced into clinical practice.

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6

PREVENTION OF PIN TRACT INFECTION IN EXTERNAL STAINLESS STEEL FIXATOR FRAMES USING ELECTRIC CURRENT IN A GOAT MODEL

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Abstract

Pin tract infections of external fixators used in orthopaedic reconstructive bone surgery are serious complications that can eventually lead to periostitis and osteomyelitis. In vitro experiments have demonstrated that bacteria adhering to stainless steel in a biofilm mode of growth detach under the influence of small electric currents, while remaining bacteria become less viable upon current application. Therefore we have investigated whether a 100 μA electric current can prevent signs of clinical infection around percutaneous pins, implanted in the tibia of goats. Three pins were inserted into the lateral right tibia of nine goats, of which one served for additional frame support. Two pins were infected with a *Staphylococcus epidermidis* strain of which one pin was subjected to electric current, while the other pin was used as control. Pin sites were examined daily. The wound electrical resistance decreased with worsening of the infection from a dry condition to a purulent stage. After 21 days, animals were sacrificed and the pins taken out. Infection developed in 89% of the control pin sites, whereas only 11% of the pin sites in the current group showed infection. These results show that infection of percutaneous pin sites of external fixators in reconstructive bone surgery can be prevented by the application of a small DC electric current

Introduction

Pin site infections of external fixators in reconstructive bone surgery frequently occur with an incidence up to 71% (Mostafavi et al. 1997, Sims and Saleh 2000) and constitute a major concern for orthopaedic surgeons. Prevention of pin site infections is also an important nursing responsibility (McKenzie 1999), but there is no consensus on how to perform optimal pin site care (Gordon et al. 2000). When a pin site becomes

infected, it is usually difficult to treat due to the formation of biofilm around the metal surface. The biofilm mode of growth shields the bacteria from the host defence mechanism and antibiotics. Literature indicates that 500-5000 times higher levels of antibiotics are needed to achieve the same antimicrobial effects on biofilm bacteria than needed for planktonic bacteria (Anwar et al. 1990, Costerton et al. 1987, Khoury et al. 1992).

The development of a biomaterial-associated infection starts with the adhesion of bacteria to the biomaterial surface, as mediated by attractive Lifshitz-Van der Waals forces, acid-base interactions and electrostatic forces (Hermansson 1999). Because all naturally occurring surfaces, including those of bacterial cells, are generally negatively charged, the electrostatic force between bacteria and a biomaterial surface is repulsive (Jucker et al. 1996). These repulsive forces can be enhanced by application of an electric current, therewith increasing the negative charge and consequently the repulsive force (Poortinga et al. 2000, Ueshima et al. 2002).

Recently, we demonstrated that it was possible to detach more than 60-76% of staphylococci adhering to surgical stainless steel surfaces through the application of small electric currents (100 μA or less), while also staphylococci growing in a biofilm could be detached through the application of an electric current (Van der Borden et al. 2004), most notably in the absence of any biocide. An electric current has been known before to enhance the bactericidal effects of many biocides, an effect called the "bioelectric effect" (Blenkinsopp et al. 1992, Costerton et al. 1994), whereas also a direct bactericidal effect of electric currents has been described (Liu et al. 1993 and 1997). Recently, we have observed this direct bactericidal effect on bacteria that remained adhering after electric current induced detachment in the absence of any antibiotics (Van der Borden et al. 2004). Note that for human application an electric current of 100 μA is well below the limit of being dangerous.

Considering the problems that infections pose in reconstructive bone surgery using external fixators, the threat posed by the ongoing (mis)use of antibiotics and the rise in antibiotic resistance amongst many human pathogens and the above described in vitro experiments, it is the goal of this paper to determine whether a direct electric current of 100 μA can prevent clinical infection around percutaneous pins, implanted in the tibia of goats.

Materials and Methods

Bacterial strains

Staphylococcus epidermidis HBH276, isolated from surface sites of premature neonate in 1990 at St. Joseph's Health Centre in London, Ontario, Canada (Bialkowska-Hobrzanska et al. 1990, Busscher et al. 1994) was used for the study after approximately 10 passages and adhered firmly and formed biofilms on different surfaces (DeJong et al. 2001). Bacteria cultured in Trypton Soya Broth (TSB, OXOID, Basingstoke, UK) at 37°C in ambient air was used for the experiments. Bacteria were inoculated from blood agar plate in a pre-culture and allowed to grow for 24 h followed by a main culture which was grown for 17 h prior to harvesting. Bacteria were centrifuged (5 min at 5000 g at 10°C) and washed twice in 10% TSB growth medium and re-suspended with 3.0×10^6 colony forming units (CFU) per ml in 10% TSB growth medium. CFU per ml was determined in triplicate prior to surgery by plate counting of a 17 h old culture and subsequently adjusted by dilution.

Electric current and electrodes

Self-drilling, self-tapping surgical stainless steel pins (5038-2-080 Apex Pin, Stryker Corp, Kiel, Germany) were used as a

cathode while a circular platinum electrode supported by a polycarbonate canister completed the circuit as an anode. The pins, connecting rods and electrodes were sterilized in an autoclave at 121^{o}C for 20 min. An aluminium housing containing a high power 9 V battery and the electronic circuit was the current source. The aluminium housing was sled over the connecting rod and connected to the negative pole of the battery. Figure 1a shows the fixation frame with the anodes and the current source attached to it. Each applied current was controlled by its own LM334Z (National Semiconductor Corp, Silicon Valley, USA), whose output potential was adapted continuously to meet the required current. A 100 µA DC current was used for the present study.

Experimental protocol

The experiments were approved by the University of Groningen Animal Ethical Committee. Nine mature female Saanen goats were used for this study. The goats were allowed free access to food and water and were unrestrained in their cages throughout the experiments. However, prior to surgery, the animals did not have any food for 8 h. Preoperatively, the animals were sedated with Thiopental (Nesdonal, 20 mg/kg i.v., AUV, Cuijk, The Netherlands), and after intubations anaesthesia was continued with a mixture of isoflurane and oxygen. Per operative Buprenorfinehydrochloride (Temgesic, 0.01 mg/kg i.v., AUV, Cuijk, The Netherlands) was given for analgesia, which was continued intramuscularly once every 24 h for 2 days to prevent postoperative pain. Furthermore analgesics were given on indication.

The right hind limb of each animal was shaved and disinfected with betadine. Incisions of approximately 1 cm were made in the skin on the lateral side of the right tibia, the first (A) 3 cm above the ankle joint and the second (B) and third (C) each 3

cm above the first and second, respectively. The 3 mm external fixation pins were inserted with the aid of a hand-drill in the far cortex of the tibia. Open wounds around pins A and B were carefully dried of blood before inoculating them with 0.1 ml of a 3 x 10^6 CFU per ml suspension of *S. epidermidis* HBH276. The bacterial suspension was pulled into the wound and in-between

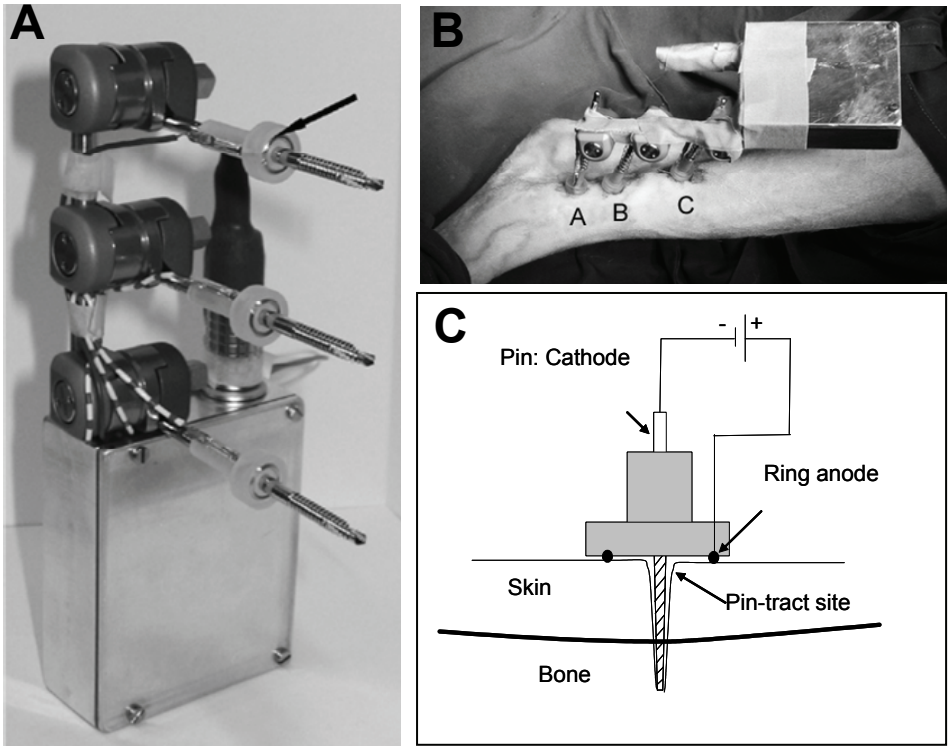

Figure 1. Current source and its placement.
(A) Photograph of the fixation frame equipped with current source. The pins act as cathodes, while the anodes are constituted by polycarbonate disks, with a platinum ring inserted to form the actual anode (arrow).
(B) Fixation frame equipped with current source and secondary electrodes implanted in the right tibia of a goat. Electric current is applied to pin A, while pin B is used as control. Pin C offers additional support to the frame and power supply.
(C) A schematic presentation illustrating the positioning of the pin in the bone, platinum electrode and the electrical connections.

the pin-skin interface by gravity. Care was taken that the bacterial suspension from one wound did not flow into the adjacent wounds. After placement of the pins and inoculation with the bacterial suspension, the sterilized anodes were

installed thus shielding and covering the wound. All pins were connected to a single rod with pin-to-rod couplings (Hoffmann II Compact, Stryker Corp, Kiel, Germany) to which the current source was attached and connected to the platinum anodes (Figure 1b). The platinum electrode around pin A received 100 µA DC current from the current source from where the current passed along the skin and wound into the pin.

Pin A was connected to the negative pole of the battery via the connecting rod and aluminium housing hence completing the circuit. The current was applied from the time of implantation until the end of the experiment on a continuous basis. Pin B was used as control (receiving no current), whereas pin C served as an additional support for the frame. Before the animals returned to their cages, the electric current and voltage were measured.

After surgery, the goats were housed individually and daily clinical evaluations of each pin site began 24 h after surgery. None of the goats were observed to lick or bite the pins. Sometimes the connecters detached and the current source displaced on the connecting rod which could be due to goats bumping into the side walls or sitting on the leg. This detachment and displacement was corrected once observed and current and voltage monitored every morning.

The infection condition of a pin was annotated as one of the following: dry pin site was considered as no infection (score 1), inflammation or moist wound (score 2) or frank purulence at the pin site (score 3) (DeJong et al. 2001). The infection was observed and judged by two independent observers, although scoring was always unanimous.

On postoperative day 21, the animals were sacrificed. From the leg with pins inserted, an X-ray photo was taken. Subsequently, to allow microscopic evaluation of the biofilms on the pins, the frame was removed and the part of the pin outside the body was whipped clean with alcohol. Next the skin and the remaining tissue were carefully dissected from each pin, to

allow removal of the pin without damaging a possibly existing biofilm. The explanted pin was submerged in staining fluid (LIVE/DEAD *Bac*light Bacterial Viability Kit, Molecular Probes, Leiden, The Netherlands) and incubated for 15 min in the dark. On different, randomly chosen locations on each surface, micrographs were taken with a confocal laser scanning microscope (CLSM) using a 40x ultra long working distance objective with the microscope set to FITC (excitation 488 nm and emission 500-600 nm) and TRITC (excitation 543 nm and emission 560-700 nm) to show dead and live bacteria, respectively.

Results and Discussion

Figure 2 summarises the infection scores for each pin site obtained by the clinical evaluations during the 21 days of the follow-up. In the control group, 1 out of 9 goats showed no signs of infection, 2 out of 9 showed inflammation and 5 out of 9 showed frank purulence. On average, inflammation occurred after 6.9 ± 3.7 days (± standard error of the mean), while at 11.7 ± 4.6 days frank purulence was observed. In the group of pins to which an electric current was applied, 8 out of the 9 pins showed no inflammation or infection. One pin out of 9 pins showed infection from the 6^{th} day onwards, which we considered unrelated to the experiment as this 9^{th} goat displayed an entirely swollen and red right hind leg with clinical signs of infection, also around the third support pin and this already within three days after surgery. The above results clearly demonstrate a clear advantage of applying electric current to prevent pin-tract infection.

Figure 3 shows X-rays of a non-infected pin A (Figure 3a) after having received an electric current and of an infected pin B (Figure 3b) in the control group. Around the infected pin there is a clear osteolytic zone visible in the bone marrow. At the

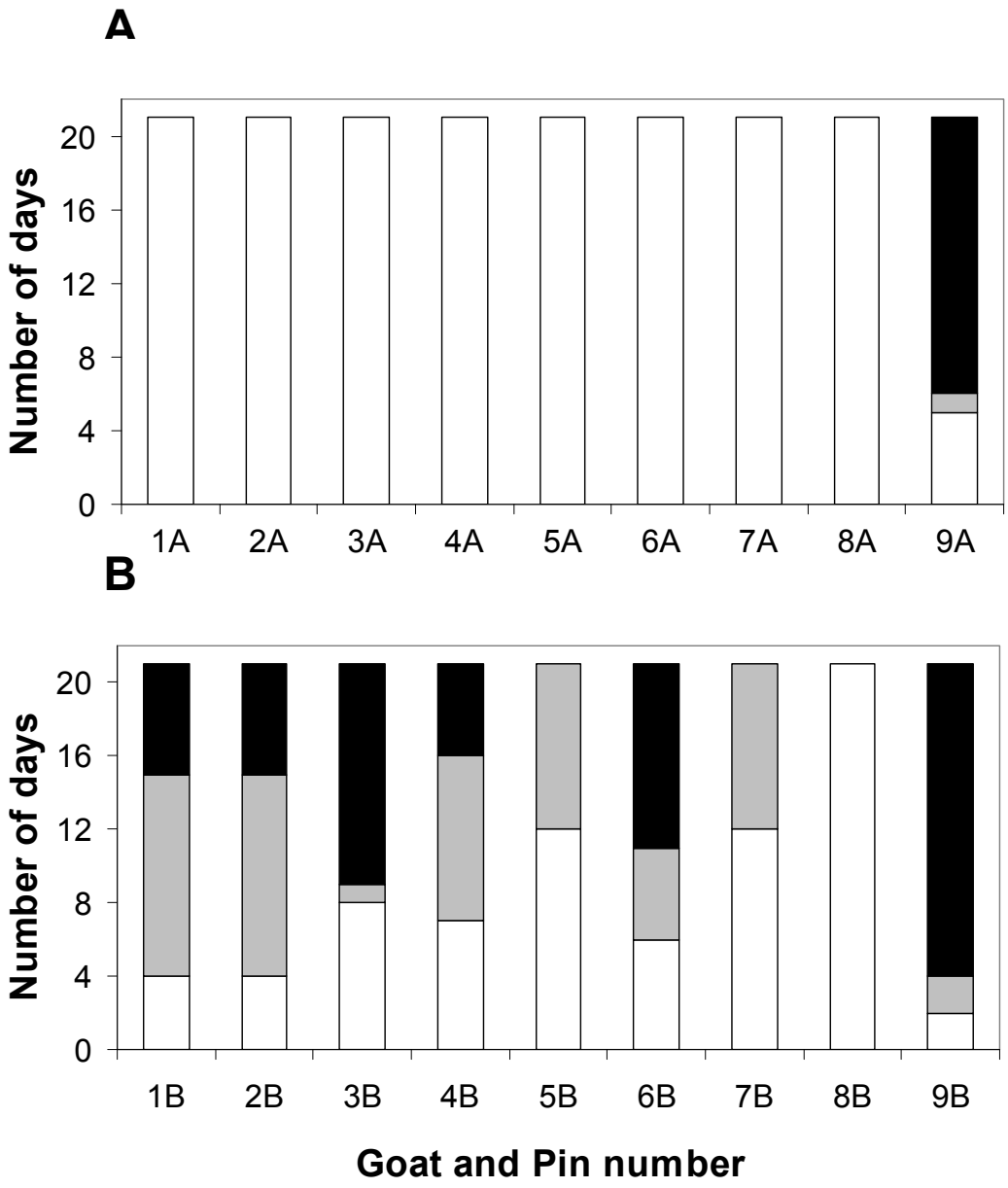

Figure 2. The development of infection over time for each implanted stainless steel pin site and goat. Intentionally infected fixation pins are grouped according to whether they receive 100 μA DC current (A) or not (B). Pin number consists of the goat number and implantation site (A or B). White indicates an uninfected pin site (score 1), grey indicates inflammation or serious drainage without frank purulence (score 2) and black indicates frank purulence at the pin site (score 3)

point of pin entry the white zone suggests reaction of the periosteum. These radiographic findings are suspect for osteomyelitis, which in this case has developed within 21 days in absence of an electric current.

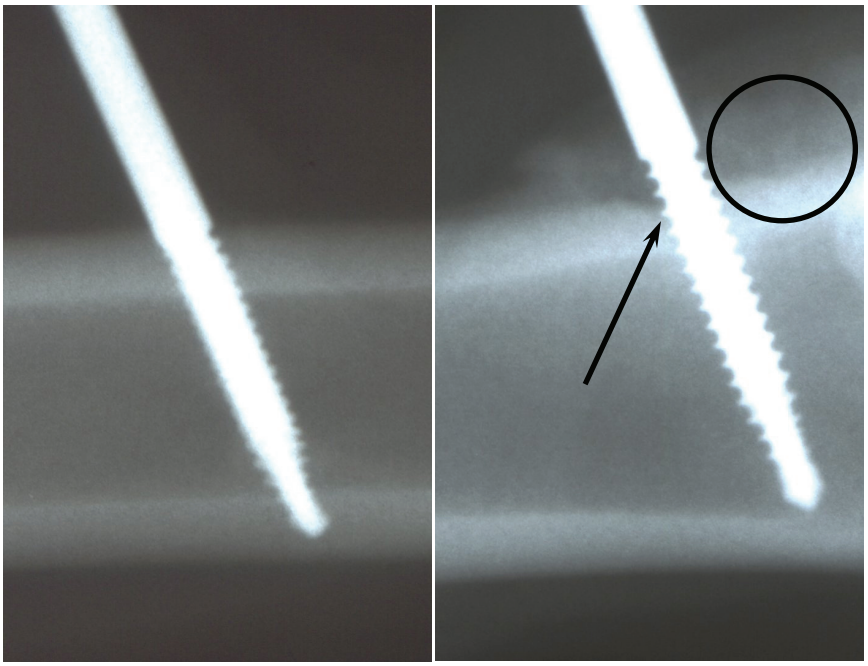

Figure 3. X-ray examples of implanted pins.

(A) A non-infected pin site, after electric current application for 21 days.
(B) An infected control pin site, in the absence of electric current application.
The inflammatory reaction of the periosteum is encircled and the osteolytic zone in the cortex is indicated by the arrow.

Figure 4 shows two confocal laser scanning micrographs after live/dead staining of a biofilm remaining on the pin's surface after removal from the animal. The applied electric current killed the majority of viable bacteria in the biofilms (Figure 4a) in comparison to the absence of an electric current (Figure 4b), while the few viable bacteria that remained evidently did not yield clinical signs of infection.

The present study tests the effectiveness of a small 100μA DC current in preventing pin-tract infection using a goat model. The effectiveness is tested in a worst case scenario where the pin-tract wound was intentionally infected with 3×10^5 CFU of *S. epidermidis* HBH276 and no additional wound cleaning steps were taken during the follow-up period of 21 days. We believe

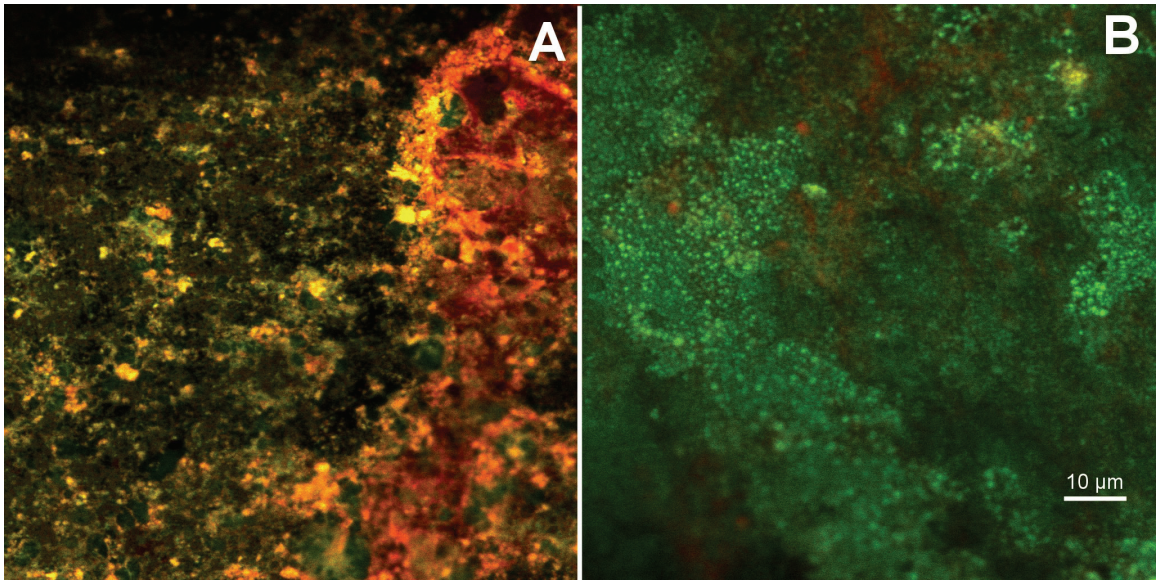

Figure 4. CLSM micrograph of biofilms adhering to a stainless steel pin after explantation from a goat.
(A) Biofilm on a pin after application of an electric current.
(B) Biofilm on a control pin, receiving no electric current.
Green represents live bacteria and red dead ones. Bar marker indicates 10 µm.

that if efficacy is demonstrated under the above conditions, electric current will also be effective in real situations where bacterial burdens are much lower and additional wound care is usually taken.

Interestingly, the method described can also be used as a diagnostic tool, as there is a clear correlation between the wound score and the electrical resistance of the skin (see Figure 5 for two examples). The wound resistance was low (~10 kΩ) directly after surgery but later as the wound dried, resistance increased. Pin 7A in the electric current group for instance, did not show clinical signs of infection and the infection score remained 1, while from day 3 onwards the electrical resistance stayed high at 6 MΩ. Pin 9A, however, became infected from day 6 on, concurrent with a drop in resistance from 6 MΩ to 25kΩ and to 10 kΩ after day 14.

The use of electric current to prevent signs of clinical infection presents major advantages in addition to or compared with

current treatment modalities. The electrodes and electric circuitry are reusable and therefore the costs can be kept low. Available fixation frames and implantation techniques can be employed without any need for modification; moreover the method does not require any antibiotics.

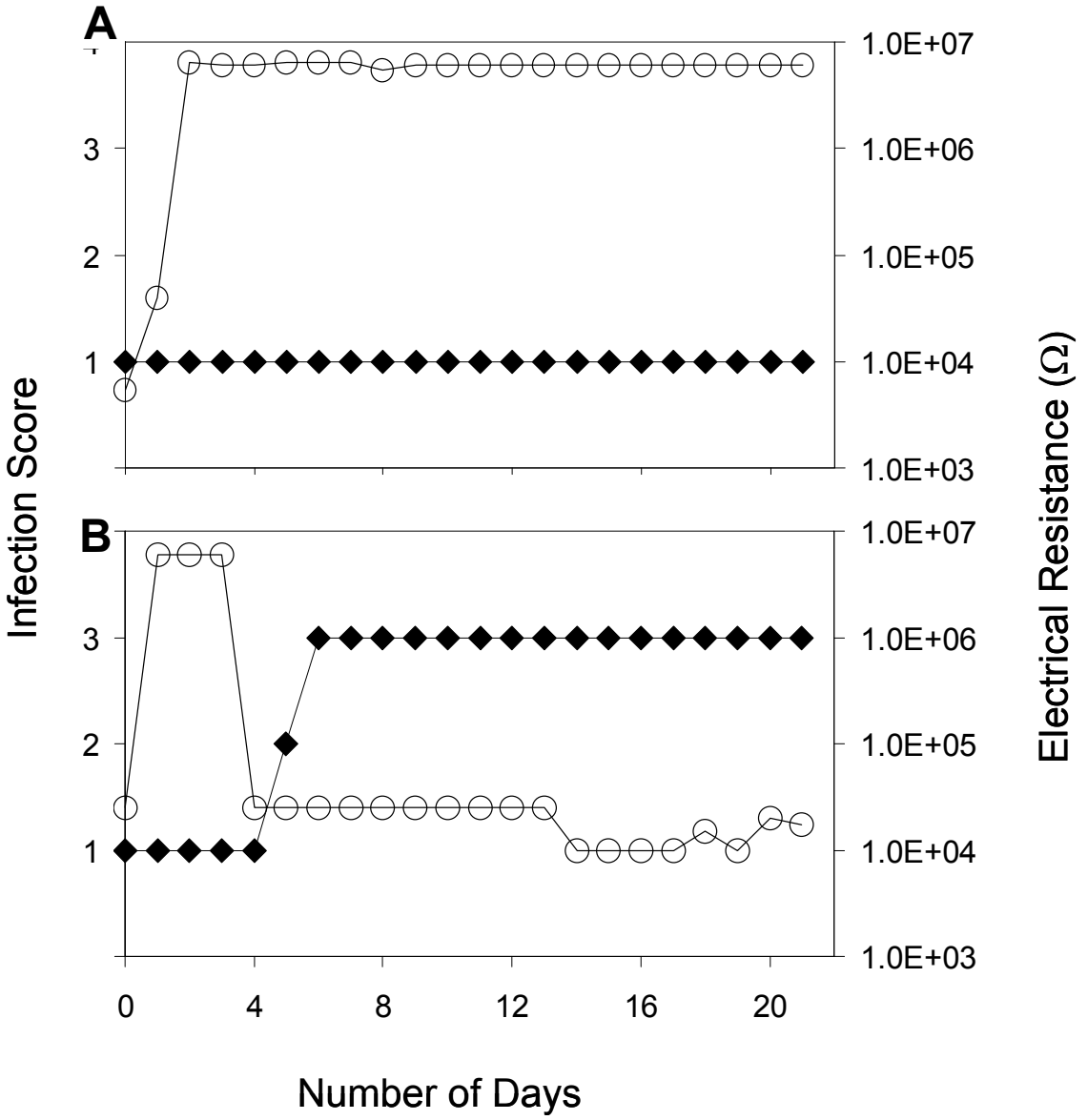

Figure 5. Infection score (closed symbols) and electrical resistance (open symbols) for pins receiving electric current as a function of time.
(A) Pin 7A, no infection developing due to current application
(B) Pin 9A, where inflammation and later infection started on the 6th day even after application of current

Conclusions

Small electric currents of 100 μA are able to prevent clinical signs of infection around surgical stainless steel pin sites, without the use of antibiotics in intentionally infected wounds, suggesting that electric currents will also be effective in real situations where the infection burden is much lower. The wound electrical resistance decreases with worsening of the infection from a dry condition to a purulent stage.

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7

TIMING OF OPEN DEBRIDEMENT FOR SUSPECTED INFECTION OF JOINT PROSTHESES
A report on 551 patients

Abstract

The decision to perform open debridement in case of a suspected acute periprosthetic infection is a most difficult one in orthopaedic implant surgery. The aim of this study is to evaluate the results of an ad hoc versus a protocolled approach, with regard to the recording of persisting wound drainage after placement of a primary joint prosthesis and the salvage of prostheses in patients with persisting wound drainage. Charts of patients after primairy total joint prostheses were studied retrospectively. 247 Patients with 250 prostheses formed group I (ad hoc approach) and were observed and treated by one of the attending orthopaedic surgeons in the absence of a protocol. In group II (protocolled group), 304 patients with 308 prostheses were observed and treated according to the proposed protocol.

The percentage of patients with a registered persisting drainage of the operative wound in group II is almost twofold that of patients with persisting wound drainage in group I (21% and 11%, respectively). Yet, the number of open debridements carried out in group II (17%) was lower than in group I (30%) and the salvage rate of prostheses with persisting drainage in group II (94%) was higher than in group I (85%). However, the main advantage was seen in the prostheses that were not debrided and remained free of infection at the last follow up, which amounted 98% in group II versus 90% in group I.

Protocolled observation and treatment yields a significant increase in the number of persistent wound drainages registered. Besides better registration, a protocolled approach enables more successful selection of patients in whom open debridement is not necessary.

Introduction

Deep periprosthetic infection following the placement of a total joint prosthesis is a major complication. Historically, in the decision-making process to deal with this complication, the infection should be classified according to onset and duration. Open debridement and retention of the infected prosthesis should be reserved for infections in the early post-operative phase. The result of this operative intervention usually eradicates between 14% and 71% of all infections (Crockarell et al. 1998, Tsukayama et al. 1996). To make early debridement successful, it should be performed within 3 to 4 weeks after the initial procedure (Hanssen and Spangehl 2004), although when the offending micro-organism is *Staphylococcus aureus* (Brandt et al. 1997) the opportunity for a successful debridement seems to be shorter than 2 days.

The decision to perform a second operative procedure is a difficult one. Every surgeon faced with this decision has to balance the probability of performing an unnecessary operative intervention with it's own chances upon complications against the probability of not performing a necessary open debridement in case of a true acute periprosthetic infection. This decision can only be made when post-operative signs are adequately noticed and reported by medical personnel directly involved in the care for these patients. In large academic centers, the medical personnel is formed by a constantly changing population with various levels of experience and knowledge, which make a proper judgement on a difficult clinical decision such as to perform open debridement extra difficult and the decision is frequently taken on an ad hoc basis. In order to maintain a high level of suspicion for cases of impending infection, we developed a protocolled approach for this clinical problem.

In order to intervene as early as possible when a periprosthetic infection is developing, persisting wound discharge is taken as a starting point in the decision protocol, which furthermore includes measurement of CRP and ESR. The presence of a superficial wound infection after placement of a total joint prosthesis has been identified as a significant risk factor for development of periprosthetic infection, but the exact extent of the risk is unknown (Abudu et al. 2002, Gaine et al. 2000, Surin et al. 1983, Knobben et al. 2006). Patients with wound discharge of 5 days or longer were reported to have 12.7 times higher risk of getting late periprosthetic infection compared to patients with a shorter wound discharge (Saleh et al. 2002). Due to the systemic effects of an operative procedure, post-operative CRP and ESR become elevated and even though these values can stay elevated for nearly a year (Aalto et al. 1984, Shih et al. 1987), CRP and ESR should show a tendency to settle down in the immediate post-operative period (Choudhry et al. 1992, Larsson et al. 1992). Persisting wound drainage after placing a primary total joint prosthesis and failure of decreasing post-operative CRP and ESR values form the basis of our decision protocol that was introduced in the orthopaedic department of our hospital in March 2003. This decision protocol is schematically presented in Figure 1.

The aim of this study is to evaluate the results of this protocolled approach with regard to the recording of persisting wound drainage after placement of a primary joint prosthesis and the salvage of prostheses in patients with persisting wound drainage. The study was carried out in a teaching hospital and a historical group of patients was used as a control (ad hoc approach).

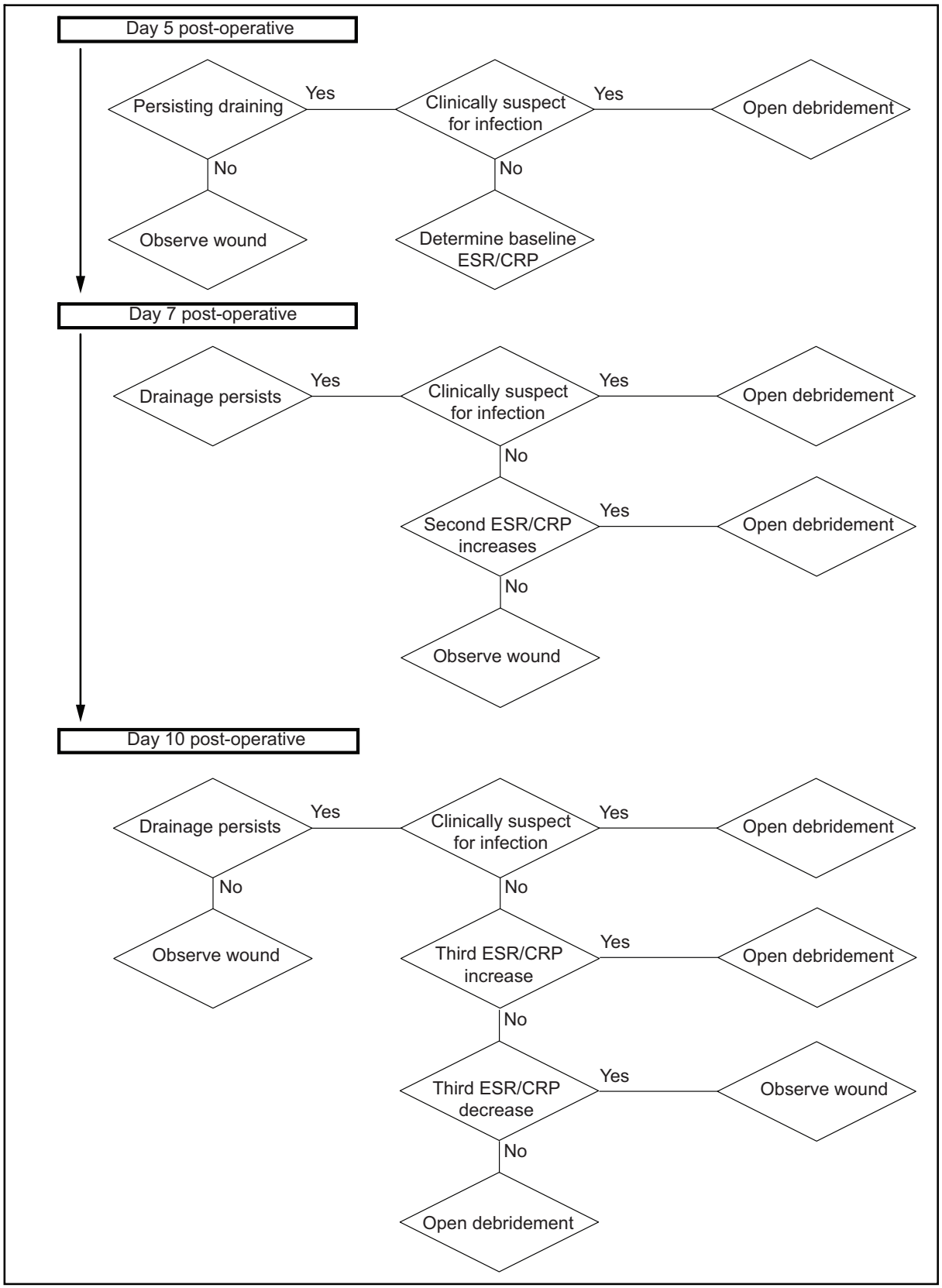

Figure 1. Decision-making protocol in case of persisting wound drainage after placement of a primary total joint prosthesis for open debridement.

Material and Methods

This retrospective study was conducted between January 2002 and January 2005. In this period a total of 558 (including 551 patients) primary total joint prostheses have been placed in the University Medical Center Groningen, of which were 192 Total Knee Arthroplasties (TKA) and 366 Total Hip Arthroplasties (THA). This study was approved by the Medical Ethical Committee for human experiments of our hospital. All operations were carried with standard peri-operative care, including 1000 mg cefazolin (Eurocept bv, Kortenhoef, The Netherlands) antibiotic prophylaxis for 24 h. For prevention of Deep Venous Thrombosis (DVT) Acenocoumarol (Centrafarm, Etten-Leur, Netherlands) 1 mg. was used. An International Normalized Ratio (INR) between 2.0 and 3.0 was considered to be adequate for preventing DVT. One deep low-pressure suction drain was used.

The study population was divided in 2 groups at the start of our protocol in March 2003:

Group I: Patients treated from January 2002 to March 2003. In this historical group, patients with a primary total joint prosthesis and a possible periprosthetic infection in the early post-operative phase were debrided by an ad hoc approach without protocol. The decision to perform an open debridement was made by the surgeon present when and if the problem was noted. This was not always the treating surgeon.

Group II: Patients treated from March 2003 to January 2005. In this group, patients with persisting wound drainage for at least 5 days were treated according to the protocol described below (see also Figure 1).

Baseline characteristics of the study population are given in Table 1. The p-values were not statistically different between the characteristics of the ad hoc group I and the protocolled group II.

Table 1. Baseline characteristics of study population.

	Group I 250 prostheses	**Group II 308 prostheses**	**Fisher's exact p-value**
Mean patient age in years	64.2 ± 13.6	66.5 ± 13.2	0.074
Males/Total (%)	67/250 (26.8%)	97/308 (31.5%)	0.302
Total Hip Arthoplasties	167	199	0.367
Total Knee Arthoplasties	83	109	0.592
Patients with reumatoid arthritis	32 (12.8%)	42 (13.6%)	0.900

Decision making process in historical "ad hoc" group

In case of prolonged drainage of the wound after placement of the primary total joint prosthesis, the decision to perform an open debridement was made by the surgeon present when and if the problem was noted. This was not always the treating surgeon. There was no clear definition of what was called prolonged drainage.

Decision making according to protocol

If persisting drainage was clear and the clinical status of the wound, according to the classical signs of rubor, calor, dolor and tumor, was not suspect for infection, baseline values of ESR and CRP were determined on day 5 and close wound observation was indicated. If these clinical signs were suspect

for infection, an open debridement was indicated. If drainage persisted until day 7, ESR and CRP were again determined. In case of persisting drainage and a suspect clinical status of the wound OR in case of an increase in ESR/CRP values, there was an indication for open debridement. If this was not the case, again close wound observation was indicated.
If drainage persisted until day 10, for the third time ESR and CRP were determined. We considered operative intervention indicated in case of persisting drainage until day 10, and only the combination of decreasing ESR/CRP values and diminishing drainage could withhold operative intervention at that moment.
During the whole postoperative period it was the surgeon's responsibility to judge the aspect of the drainage and/or clinical status of the wound in order to decide for or against open debridement.

Post-operative treatment in case of open debridement performed

In both groups, the treating surgeon decided intra-operatively, based on the macroscopic aspect of the periprosthetic tissue on the treatment modality to be used. If infection was not clearly present, biodegradable gentamycine-fleeces (GentaFleece®, Baxter AG, Vienna, Austria) were used. In cases of macroscopic pus and/or clearly inflamed periprosthetic tissue, gentamycine-beads (Septopal®, Merck KGaA, Darmstadt, Germany) were placed. Removal of the gentamycine-beads necessitated a second opening of the wound after 2 weeks, during which re-debridement was carried out when deemed necessary. After at least five samples had been taken from different places of the peri-prosthetic tissue during the first open debridement, combination antibiotic therapy was started until the culture results were known (Flucloxacilline (GlaxoSmithKline BV, Zeist, the Netherlands) 4 dd 1000 mg intravenously (iv) +

Rifampicine (Aventis Pharma BV, Hoevelaken, The Netherlands) 1 dd 600 mg iv). As soon as the infecting organism was identified, antibiotic therapy was changed concordant to the antibiogram for a total of 6 weeks. Otherwise, the combination antibiotic therapy of flucloxacilline and rifampicine was maintained for 2 weeks intravenously and changed to an oral regime in the same dosage, to complete a total of 6 weeks of antibiotic therapy.

Periprosthetic infection was defined as growth of identical micro-organisms on cultures of three or more specimens out of at least five specimens obtained during open debridement (Atkins et al. 1998). All patients were followed postoperatively with respect to persisting wound drainage.

Failure of treatment was defined as a revision of the primary prosthesis because of septic complications. For the statistical analysis of the data, Fisher's Exact test was used.

Results

During the study period, 91 out of 551 patients had a persisting post-operative drainage of the wound (see Table 2): 27 patients out of 247 (11%) in the ad hoc group I and 64 out of 304 (21%) in the protocolled group II, which is a significant difference ($p = 0.0002$).

In group I, 8 out of 27 cases were treated with an open debridement. In this group there were 4 patients with a TKA and 4 with a THA. Among the 19 patients (70%) that were not debrided, 5 patients had a TKA and 14 had a THA. In group II, open debridement was performed in 11 out of 64 cases. Although the percentage of open debridements performed in group II was clearly smaller compared to group I, 17% vs. 30% respectively, this was not a significant difference ($p = 0.258$). Among the debrided patients in group II, there were 4 patients

with a TKA and 7 with a THA and among the non debrided there were 25 patients with a TKA and 28 with a THA (83%), all showing persistent wound drainage. In group I the mean time from the initial procedure until the moment of open debridement was 14 days (7-22 days), whereas in group II this was 10 days (6-16 days).

Table 2. Registration of persistent wound drainage, number of open debridements carried out and their outcome with respect to revision or salvage of the prosthesis in group I (treated ad hoc) and group 2 (treated according to protocol).

	Group I N=247 patients		**Group II N=304 patients**	
Patients with persistent wound drainage	27		64	
	Open debridement carried out	No debridement carried out	Open debridement carried out	No debridement carried out
Persistent wound drainage	8	19	11	53
Prostheses revised	2	2	3	1
Total number of revised prostheses	4		4	
Total number of salvaged prostheses	23		60	

The number of failures of open debridement in group I, 2 out of 8 (25%), and in group II, 3 out of 11 (27%) were not statistically different (p = 1.00). Group I and group II both had a total number of 4 failed prostheses. For the total group of patients with persisting drainage, the number of salvaged total joint prostheses increased from 85% (23 of 27 patients) in group I to 94% (60 out of 64 patients) in group II. In group I, 2 out of 19 had a two-stage revision because of septic complications, while in group II only 1 out of 53 patients had a two-stage revision, which is not significant.

Table 3. Overview of cultured micro-organism from the operatively debrided prostheses in group I and group II.

Group I	
Micro-organism cultured	Isolation frequency
Staphylococcus aureus	2
Coagulase-negative staphylococci	4
Enterococci	1
Unknown	2
Group II	
Staphylococcus aureus	5
Coagulase-negative staphylococci	2
Klebsiella pneumoniae	1
Pseudomonas	3
Candida albicans	1
Unknown	1

In the combined two groups, 19 open debridements were carried out and in 16 patients infecting organisms could be determined (see Table 3). In the majority of these patients only

one organism was cultured. In 3 patients there were two infecting organism, of which one was always *S. aureus*. Two patients in group I did not harvest any micro-organisms, while in only one patient of group II infection could not be microbiologically confirmed.
In the study population of 551 patients with 558 prostheses, an overall total of 11 (2.0%) have been revised because of septic complications. This number includes the revised prostheses from the persisting drainage group and the two-stage revision of prostheses that did not have prolonged drainage from the operative wound.

Discussion

In a large study population, we found our protocolled observation and treatment of patients with persisting drainage of the wound after a primary total joint prosthesis to be successful in electing patients in which an open debridement was not necessary. We regard the study population of 551 patients as representative, considering the characteristics of this population (compare Table 1) and the micro-organism cultured after open debridement (see Table 3). Also the percentages of revised prostheses due to septic complications (2.0%) are comparable with other studies (Hanssen and Rand 1999, Wymenga et al. 1992).
Several authors have stressed the importance of the elapsed time between the index procedure and the moment of open debridement in case of a suspected early deep prosthetic infection of a TKP (Deirmengian et al. 2003, Mont et al. 1997, Rand 1993, Silva et al. 2002). All agree that the sooner open debridement is performed, the higher the chances of prosthesis salvage become. However, literature offers little help in the difficult decision as to whether it is safe to continue observation or even withhold open debridement in case of persisting

drainage after primary total joint prosthesis. In our study, fewer revisions in the absence of open debridement were performed in case persisting drainage was monitored according to a protocol. Although this change is clinically very important, this statement could unfortunately not be made statistically significant at a high p-value, despite the large initial numbers involved in this study.

Our protocol "persisting wound drainage after primary total joint prosthesis" dictates the timing for debridement in case of a persisting drainage after placement of a primary total joint prosthesis. The decision has to be made within the time frame of the fifth until the tenth day after the primary procedure. The clinical aspect of the operative wound and the aspect of drainage are the key factors in the decision to perform or withhold an open debridement. Only a decrease in ESR and CRP or a decrease in drainage can postpone or alter this decision. By following this protocol the elapsed time from the index procedure to a debridement has diminished from a mean of 14 to a mean of 10 days.

Application of the protocol led to a salvage percentage of 75% in the debrided group, which was not significantly different from the ad hoc group 73%. Apparently, there is window of opportunity for successful salvage of an acute postoperative prosthetic infection. For early open debridement to be successful, it should be performed within 3 to 4 weeks after the initial procedure (Hanssen and Spangehl 2004). This study shows that there is no benefit of performing an operative debridement after a mean of 10 days instead of 14 days.

After the introduction of the protocol, there seemed to be an increase in the percentage of patients with persisting drainage of the operative wound after placement of a primary total joint prosthesis and this can probably be explained by a raised awareness among the staff due to the introduction of the protocol. Despite this increase in the number of patients with a registered persisting drainage, the number of open

debridements decreased in this group. The decision modifying parameters of increasing or decreasing levels of ESR and CRP appear to control the number of debridements and provide a guideline to the treating surgeon with which he feels comfortable. Note, that this may be especially true in our large teaching hospital. Moreover, microbiological confirmation of infection could not be obtained in two patients of group I and in one patient of group II, which is an additional argument in favour of a protocolled approach. One drawback in the present protocol is the fact that in this use of post-operative levels of ESR and CRP it is sometimes hard to ascertain what magnitude of increase or decrease in these levels is to be considered significant. Another problem we encountered during the research period is that the judging of the clinical aspect of the wound based on the classical signs of infection, the judging of the aspect of drainage and the amount of drainage was subjective. These subjective judgements and their large impact on the decision to perform a secondary operative procedure probably have influenced our results to some extent.

The mean follow up period was 2.75 years, which might be considered short in the eyes of some (Mont et al. 1997, Schoifet and Morrey 1990). However, a deep prosthetic infection can develop anytime and a follow up time of almost three years as in this study must be considered sufficient to conclude on possible benefits of a protocolled versus an ad hoc approach.

To our knowledge this is the first protocol in literature that seems capable to successfully elect patients in which it is safe to withhold an open debridement in case of persisting drainage in the early post operative phase after placement of a primary total joint prosthesis. Successful election was evidenced by a lower number of open debridements and a higher number of salvaged prostheses in patients not debrided, which is apart of cost savings and the prevention of major patients discomfort due to revision surgery.

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8

GENERAL DISCUSSION

Introduction

There are many factors affecting a patient's risk for infection during surgical procedures and even the tiniest effort to minimize postoperative infections and subsequent suffering of the patients involved can be important to reduce Surgical Site Infection (SSI). Unacceptably high infection scores on our orthopaedic ward prompted us to investigate possible causes and to evaluate contamination problems in total joint replacement surgery and ascertain possible causes of these high infection rates. The aim of this thesis was to investigate the reasons for diagnostic problems in a University Hospital setting in total joint prostheses suspected of infection. Diagnostic problems are analyzed during the work up for revision surgery as well as during the peri-operative hospital stay after primary hip replacement. A possible method of preventing clinical signs of infection of a percutaneous orthopaedic implant, which is even more susceptible to infection than totally internal implants, was investigated in an animal model.

Surprisingly during the course of this research, we found a clear change in attitude among all those involved in patient care at different levels towards pre-operative and peri-operative attitude in biomaterial-associated infection problems.

The basis of this change of attitude is thought to be the "Hawthorne effect" (See Highlight). In several fields of science worldwide it can be seen that research is a valuable tool to create greater awareness and stimulate changes in a broader society than solely those involved with research. For instance, dental health in Japan is far behind compared to Western-Europe and the United States, where the pathway toward improvement has been established in the past and the general public knows the value of preventive measures (Kawamura and Iwamoto 1999). Yet, in order to implement the use of such

preventive programs elsewhere, research has to be conducted at universities and industries to create an awareness of the problems and its solutions among researchers, professionals and finally the general public in order to facilitate real changes.

They Call it the Hawthorne Effect

In the 1930's some studies were held at the Western Electric production facility outside Chicago in a place called Hawthorne. The intent of the study was simple enough: invite a handful of employees to participate in various working condition tests to determine which conditions were most conducive to increased production. Those conditions that "tested" best were then to be rolled out to the general production floor. One of things they tested was brighter lights. Production went up. Then they tested dimmer lights. Production went up. In fact, no matter what they tested, production went up!

By singling out a small group of employees to participate in an exclusive trial, participants felt valued, special and important. The special attention they received gratified their ego and created a positive emotional bond with what they were trialling. The practical upshot was that the research trials effectively transformed the research participants into advocates for whatever it was they were trialling.

This is despite the fact that the outcome of the research is known in a sense (Hugoson et al. 2005, Okawa et al. 1992). The virtues of such an approach toward decreasing the infection scores at our orthopaedic ward became clear from the on-set of the study described in Chapter 3. During this study, data were collected concerning per-operative contamination of instruments used during total hip and knee arthroplasties. In the early parts of the study, surgeons frequently inquired “how did I do” and they were proud when their instruments were not contaminated during surgery. Soon, it was noticed that after a surgeon had received bad news once or twice about his or her

performance, per-operative contamination of his or her instrumentation disappeared. As soon as we noticed this, we stopped giving feedback to the surgeons about their performance (and the previously observed pattern of peri-operative contamination re-appeared). However, this incident clearly shows how research can stimulate behavioural changes in the right direction.

In the remainder of this general discussion, we will discuss the off-spring in terms of changes brought about in the treatment of patients by the research conducted at UMCG in collaboration between the departments of Orthopaedic Surgery and BioMedical Engineering, University Medical Center Groningen, Groningen, where the control of biomaterial-related infections is a topic area, on infection in orthopaedics in general, and subsequently we will focus on changes established in diagnostic methods and peri-operative attitude.

The most important clinical implication of the thesis of Dr. Hilbrand van de Belt (2001) was the realisation that micro-organisms in septic complications after total joint arthroplasty can survive treatment even when antibiotic-loaded bone cement is used as a therapeutic means.

Because of the lack of adequate control of the release of antibiotics from bone cement that might contribute to the development of antibiotic resistance among the infecting organisms, we postponed the use of antibiotic-loaded bone cement in primary total joint arthroplasties at that time until more evidence became available that would support its use in primary total joint arthroplasty next to its use in revision surgery.

Scientifically, this thesis formed the start of extensive implant retrieval studies. These retrieval studies formed the base of our knowledge of biomaterial-associated infections, as currently existing in our Orthopaedic department.

After the thesis of Dr. Danielle Neut (2003), we realised that the standard hospital culturing regime was too short for

adequate diagnosis. As a result of this thesis, and in close communication with the hospital Microbiology Department, the culturing time of implant related infections was prolonged to 3 weeks. A specific protocol concerning the number and way of tissue cultures to be taken was introduced (Atkins et al. 1998). The use of swabs was discarded and at least five tissue samples were taken from the periprosthetic tissue.

The thesis from Dr. Hans Hendriks (2003) showed that the initial burst release of antibiotics from antibiotic loaded bone cement is caused by the dissolution of antibiotic particles that are readily available on the surface of the bone cement. The long-term release appeared to be the result of the water very slowly entering the polymer matrix and carrying the antibiotic to the surface. This release of antibiotics (gentamycin) could be enhanced by the application of ultra-sound, possibly by micro-streaming or localized temperature rise.

These data together with the data published in the Swedish and Norwegian Hip Register (Havelin et al. 2000, Malchau et al. 2002), yielded the decision to also use antibiotic-loaded bone cement for primary total joint arthroplasties.

The results of the thesis of Dr. Arnout van der Borden (2005) is still part of ongoing research on preventing and/or curing infection of percutaneous metal implants through the application of small electric currents. He showed that small electric currents ranging from 15 to 125 μA stimulated various staphylococcal species to detach from stainless steel. In a pilot in vivo study, it turned out to be possible to prevent and treat infection on percutaneous pins in the tibia of goats.

The results of the thesis of Dr. Geert Ensing (2006) showed that in in vitro experiments, the release of gentamycin from gentamycin-loaded beads could be significantly enhanced by the application of ultra-sound. Moreover, organisms were found to be more susceptible to antibiotics during application of ultra-sound. This enhanced antibiotic release and efficacy was confirmed in in vivo experiments in animals. The results of

these animal experiments have promising additional value in possible clinical situations, e.g. by using ultrasound in conjunction with gentamicin beads in two-stage revision surgery in septically failed arthroplasties.
Although this thesis not yet has had a direct visible clinical impact, it has stimulated creative thinking among all those involved in patient care on how to perform "better".
The thesis of Dr. Bas Knobben (2006) clearly underlines the importance of strict behavioural- and systemic measures in decreasing intra-operative bacterial contamination. An intra-operative bacterial contamination decrease from 34% to 9% was found in his study after re-introducing behavioural measures and installation of an improved laminar flow system in our Operating Room (OR). In his study on intra-operative contamination, he found a significant association between this contamination and prolonged wound discharge.
The impact of this thesis on diagnosing biomaterial-associated infection at the orthopaedic ward-UMCG became clear to us during our retrieval study. It was noted that in the beginning of the study several loosened total joint prostheses were deemed to be aseptically loosened, while in our research laboratory viable micro-organism were cultured in 60% of the cases. These findings were shared with the surgeons in the early parts of the study and during the course of the study, without explicit changes in the OR settings, the number of retrieved truly a-septically loosened prostheses inclined. We strongly believe this phenomenon can be explained by the "Hawthorne-effect" as well. By giving feedback of the study results surgeons involved became more aware of the importance of their pre-operative workup in revision arthroplasty surgery and optimized their attitude. This positive change is reflected in the following analysis of the pre-operative workup of one-stage aseptically loosened Total Hip Arthroplasties (THA) in two separate periods, June 1999 to May 2001 (old situation) and October 2003 to January 2005 (new situation)

Pre-operative workup (old situation)

Considering the workup for the revision procedure pre-operative Erythrocyte Sedimentation Rate (ESR) and C-reactive protein (CRP) levels were determined. In Group I the mean pre-operative value of Erythrocyte Sedimentation Rate (ESR) was 17 mm/h (1-51) and the mean pre-operative C-reactive protein (CRP) level was 7 mg/l (3-24). In 12 out of 20 patients these serum levels were tested.
In group I, ^{99}Technetium bone scanning as workup was performed in 4 out of 20 cases. In only one case, ^{111}Indium leukocyte scanning was additionally performed.

Pre-operative workup (new situation)

The mean pre-operative value of ESR was 8 mm/h (2-22) and the mean pre-operative CRP level was 6 mg/l (3-16). In 14 out of 15 patients these serum levels were tested.
^{99}Technetium bone scanning as workup was performed in 8 out of 15 cases. ^{111}Indium leukocyte scanning was additionally performed in 2 cases.
This change in pre-operative workup shows that through research performed on presumed aseptical loosening of total joint prostheses, the index of suspicion of the treating surgeons shifted towards an attitude that loosening is low-grade septic unless proven otherwise.
Of course the diagnosis of periprosthetic infection is hampered by the low metabolism of the organisms involved. Even more sophisticated diagnostic tools as 18FDG-Positron Emission Tomography, although promising as has been shown in Chapter 5, have not yet given us the 100% sensitivity and specificity one would hope for.

Despite the apparent distinction between the two ways of loosening of a joint prosthesis much debate remains to what extent aseptic loosening is truly aseptic. The fact that there is no current consensus in the orthopaedic surgery or infectious diseases communities, as to what constitutes definitive evidence of prosthetic joint infection, makes the distinction between aseptic and septic loosening not easy. Unfortunately, the main symptom of both aetiologies, joint prosthesis dysfunction, is similar.

Accurate diagnosis requires the use of a combination of tests and a strong clinical suspicion. The use of more sensitive tests in an attempt to identify every clinically important micro-organism on the prosthesis or in peri-prosthetic tissue will increase the number of unnecessary two-stage revision procedures because of the presence of necrotic bacteria or contaminants (Mariani et al. 1996, Tunney et al. 1999). Possibly the potential clinical importance of endotoxins in aseptic loosening (Akisue et al. 2002, Bi et al. 2001, Ragab et al. 1999) and/or periprosthetic infection is an explanation of this transition zone between these aetiologies, but requires further study.

Until this time, the high index of suspicion of the treating orthopaedic surgeon stays of paramount importance. This suspicion however, needs to be discussed in close communication between the surgeon, the microbiologist and pathologist to couple the different test results to the appropriate clinical setting.

The impact of this thesis on the peri-operative behaviour in biomaterial-associated surgery in relation to infection at the orthopaedic ward-UMCG became clear during our study on timing of debridement. In case of persisting drainage after primary total joint prosthesis, we realized that in large academic centres, the medical personnel is formed by a constantly changing population with various levels of experience and knowledge. This makes a proper judgement on

some clinical decisions difficult and the decisions taken tend to be on an ad hoc basis. In order to maintain high levels of knowledge and experience, a protocolled approach turned out to result in improved clinical practice. A protocolled approach could be applicable in various other areas of clinical decision making as well.

Of course it is not realistic to relate every positive change in the peri-operative behaviour on our ward solely to this thesis. One major factor contributing to the improved situation must also be attributed to the infectious complication registration in 1996, as part of a larger project called PREZIES ("PREventie ZIEkenhuisinfecties door Surveillance"), in translation "Prevention of Hospital infections by Surveillance". This project develops implements and exploits a surveillance system for hospital infections. It was further developed into the "CHIPS"-project (CHIrurgische Profylaxe en Surveillance), in translation "Surgical Prophylaxis and Surveillance", and the "Doorbraak" or "Breakthrough"-project. The CHIPS-project is an intervention study on the quality and efficacy in the use of antimicrobial means in surgical prophylaxis in hospitals in the Netherlands. The "Doorbraak"-project has started in May 2003 and aims at updating pre-, peri- and post-operative procedures to minimize SSI.

In this project several events in the chain of possible contamination of total joint prostheses during the peri-operative phase have been studied and adapted to modern standards. Examples of these events are; the number of people present during total joint surgery, the number of OR door-movements when total joint surgery is performed, the timing of administration of antibiotic prophylaxis. After evaluating this project over the time period of October 2003 until January 2005, the results were published in July 2006 (PREZIES registration, UMCG 2006). These results show that the total number of SSI in total joint surgery had decreased from 9.5% (September 2001- October 2003) to a preliminary percentage

of 2.4 for 2005 (Chi^2 $p<0.005$, although not all patients had been followed for one year at the time of printing). For deep SSI this percentage decreased from 4.9 to 2.1 respectively.
One should take into account as well the fact that the morbidity of the patient population differs in a University Hospital from the one in non-academic hospitals. Recent research in our University Hospital confirms this fact (Manuscript accepted, pending revision). Accounting for this co-morbidity into the expected infection percentages for the period of October 2003 until January 2005 for Total Hip surgery, one would expect an infection percentage of 3.9%. The registered percentage was 2.9%.
Concerning the prevention and cure of infections of percutaneous orthopaedic implants, we conducted an animal study based on the results of the pilot study of Dr. van der Borden. In this study it turned out to be possible to prevent and treat infection of percutaneous orthopaedic implants in the tibia of goats. Because of the promising results in these animal experiments (Chapter 6), human studies will be set up in the near future. Further research towards preventing and treating infections in totally implanted orthopaedic implants as Total Hip Arthroplasties or Total Knee Arthroplasties are possibilities that have to be further investigated.

Summary and conclusions

The concluding question to be addressed here is: "Has our clinical practice towards the care for patients with total joint prostheses benefited from the research described here?"
Considering the changes that have been achieved during the course of this study, the answer has to be a definitive "YES!!!" Our clinical care for patients with a total joint prosthesis with regard to infection has become more based on biomaterial-associated surgery consensus in our large academic hospital

setting. This improved clinical care is found in various parts of our treatment of patients with a total joint prosthesis, ranging from pre-operative work-up to observations according to a protocol and treatment of problems on our orthopaedic ward:
Our pre-operative workup is more based on the assumption that every loosened prosthesis is considered to be a septic one and therefore the index of suspicion has been raised.
Our post-operative observations have been standardized and improved, providing the means for optimal decision making in a medical health personnel with various levels of experience.
Our total incidence rate of deep infections in total joint surgery has decreased from 4.9 to 2.1%.
Finally it should be noted that the improvements found are the result of several smaller and larger contributions from different people involved in various parts of the process. It is the result of team effort.
This study, as well as others, shows that performing research on a clinically important problem will not only result in a gain on knowledge on the issue, nor should a gain of knowledge be the sole goal of research. It also illustrates that the so-called "Hawthorne-effect", can be an effective means in optimizing behaviour of personnel involved by creating a greater awareness and stimulating changes in a broader community than solely those involved with research. This should be considered equally important on a local level than world-wide dissemination of knowledge.
The possibility of preventing or even treating an infection of a percutaneous orthopaedic implant using a small electric current is very attractive because of the ease of it's application without, until now, no side-effects shown. Of course this method is still in an experimental phase but future human experiments will show us whether this method can be extended towards totally implanted orthopaedic implants.

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9

SUMMARY

Summary

In Chapter 1 a general introduction has been given to the problem of biomaterial-related infection in orthopaedic surgery. The aim of this thesis is formulated as to investigate the reasons for diagnostic problems of infection in total joint prostheses in a University hospital setting. Diagnostic problems are analyzed during the work up for revision surgery as well as during the peri-operative hospital stay after primary hip replacement. A possible method of preventing clinical signs of infection of a percutaneous orthopaedic implant, which is even more susceptible to infection than totally internal implants, is investigated in an animal model.

In Chapter 2 an overview has been given of biomaterial-related infections and why they need special attention. Special attention to minimize contamination risks has to be brought to the attention of non biomaterial-associated Operating Room (OR) personnel. The problem we encountered in this particular part was the difficulty to maintain this attention at a high level. The latter is especially a problem in our specific setting, a large academic hospital, in which there is a constant rotation of OR personnel in training. It turned out to be of the utmost importance that the constant factors, e.g. the dedicated orthopaedic scrub team together with our orthopaedic staff, worked together to keep OR personnel focused on the specific biomaterial–associated OR attitude.

The objective of the study in Chapter 3 was to detect possible bacterial contamination in total hip arthroplasty through instruments used at the direct site of implantation during the primary procedure. In this study samples of the broaches used for preparing the acetabulum and femur, as well as samples of the reamed acetabular and femoral bone, were collected during

67 consecutive primary total hip arthroplasties in 67 patients. Broach samples were taken at the start and end of every reaming procedure. The total number of samples taken amounted to 402, of which 26 were found to be positive for micro-organisms. In 20 patients, at least one of these positive samples had been in direct contact with the actual prosthesis site, indicating that at least 30% of the involved patients had a possible bacterial contamination when leaving the operating theater.

The objective of the study in Chapter 4 was to describe the extended culturing method that has been developed at the Department of BioMedical Engineering of our University. This method has been used for culturing tissue excised in revision surgery for septic as well as presumed aseptic loosing of joint prostheses. We have retrieved 33 prostheses or prosthetic components. In only one of the 33 cases micro-organisms were detected by routine culturing of tissue, while extensive culturing demonstrated infectious organisms in tissue samples in 14 cases. In addition, extensive culturing of the biomaterial scrapings identified 6 other cases of positive cultures, totalling the percentage of infected cases by extensive culturing to 60% (20 out of 33 patients). These results demonstrated that biomaterial-associated infections may well remain undetected by standard clinical and microbiological hospital procedures

The objective of the study in Chapter 5 was to evaluate the feasibility of 2-[^{18}F]fluoro-2-deoxy-D-glucose-Positron emission tomography (FDG-PET) for the differentiation between septic and aseptic loosening of joint prostheses. Seven patients with a painful joint prosthesis had a revision procedure of this prosthesis within 6 weeks after PET scanning. Four patients had an intense FDG uptake. At revision, 3 patients were diagnosed as septically loosened, and one had an infectious-like mass around the stem. The other three had mild activity and were

diagnosed as aseptically loosened. Five control patients with 7 asymptomatic joint prostheses went through a PET scan for an oncological problem. The PET activity varied between no (N=1), mild (N=5) and intense (N=1). We concluded that the introduction of a joint prosthesis apparently causes a mild increased FDG uptake, suggesting the evocation of a chronic inflammation. An intense uptake of FDG could be suggestive of a summation of activity from a possible infectious origin.

The objective of the study in Chapter 6 was to investigate whether a 100 μA electric current can prevent signs of clinical infection around percutaneous pins, implanted in the tibia of goats. Three pins were inserted into the lateral right tibia of nine goats, of which one served for additional frame support. Two pins were infected with a *Staphylococcus epidermidis* strain of which one pin was subjected to electric current, while the other pin was used as control. Pin sites were examined daily. The wound electrical resistance decreased with worsening of the infection from a dry condition to a purulent stage. After 21 days, animals were sacrificed and the pins taken out. Infection developed in 89% of the control pin sites, whereas only 11% the pin sites in the current group showed infection. These results show that infection of percutaneous pin sites of external fixators in reconstructive bone surgery can be prevented by the application of a small DC electric current.

The objective of the study in Chapter 7 was to evaluate the results of an ad hoc versus a protocolled approach, with regard to the recording of persisting wound drainage after placement of a primary joint prosthesis and the salvage of prostheses in patients with persisting wound drainage. In this study, 247 patients with 250 prostheses formed group I (ad hoc approach) and were observed and treated by an orthopaedic surgeon in the absence of a protocol. In group II (protocolled group), 304

patients with 308 prostheses were observed and treated according to the proposed protocol.
The percentage of patients with a registered persisting drainage of the operative wound in group II was almost twofold the percentage of group I (21% and 11%, respectively). Yet, the number of open debridements carried out in group II (17%) was lower than in group I (30%). The salvage rate of prostheses with persisting drainage in group II (94%) was higher than in group I (85%). However, the main advantage was seen in the percentage of salvaged prostheses which were not debrided and amounted 98% in group II versus 90% in group I.
Besides better registration, a protocolled approach enables more successful election of patients in which open debridement is not necessary.

The general discussion in Chapter 8 illustrates that the so-called "Hawthorne-effect", can be an effective means in optimizing behaviour of personnel involved in the chain of care for patients with an orthopaedic implant. This optimized behaviour is realized by creating a greater awareness and stimulating changes in a broader community. This community is expanded beyond those solely involved with research. This kind of research is considered to be equally important on a local level than world-wide dissemination of knowledge.

SAMENVATTING

Samenvatting

In hoofdstuk 1 wordt een algemene introductie gegeven omtrent biomateriaal-gerelateerde infecties in de Orthopedie. Als doelstelling van dit proefschrift wordt geformuleerd het onderzoeken van de moeilijkheden bij het stellen van de diagnose van een infectie bij een gewrichtsprothese in een Universitair Medisch Centrum. Deze moeilijkheden worden geanalyseerd ten tijde van de voorbereiding van een eventuele revisie operatie, als ook tijdens het ziekenhuisverblijf in de periode rondom de plaatsing van een gewichtsprothese.
Verder wordt in een dier model de mogelijkheid onderzocht om te voorkomen dat bij door de huid (percutaan) geplaatste orthopedische implantaten zich een klinische infectie ontwikkelt.

In hoofdstuk 2 wordt een overzicht gegeven van biomateriaal-gerelateerde infecties. Centraal gegeven van dit type infecties is het feit dat de betrokken micro-organismen een soort "slijmlaag" vormen op een biomateriaal oppervlak, een zogenaamde "biofilm". Deze biofilm beschermt de bacteriën tegen o.a. het immuunsysteem en antibiotica, waardoor de infectie veel moeilijker te behandelen is. Biomateriaal-gerelateerde infecties zijn hierdoor niet vergelijkbaar met andere infecties, zoals een geïnfecteerde wond na een ingreep aan buikorganen. Een infectie met biofilm bacteriën maakt dat over het algemeen het biomateriaal, zoals in de Orthopedie bijvoorbeeld de gewrichtsprothesen, verwijderd moet worden om de infectie te behandelen. Met name de noodzakelijke speciale aandacht om deze complicatie te voorkomen, wordt middels een literatuur overzicht aangegeven. Speciale aandacht is nodig om de contaminatie risico's op de Operatie Kamer (OK) te minimaliseren en onder de aandacht te brengen van OK-personeel, dat niet gewend is aan de speciale maatregelen die het voorkomen van biomateriaal-gerelateerde infecties

probleem vraagt. Een belangrijk probleem dat wij in het hele onderzoek tegenkwamen, was de moeilijkheid om de aandacht voor het maximaal verkleinen van deze contaminatierisico's op een gewenst hoog niveau te blijven houden. Dit bleek vooral een probleem in onze specifieke omgeving; een groot academisch ziekenhuis, waarbij er veel roulatie is van personeel in opleiding. Het bleek uiteindelijk van groot belang dat de stabiele factoren, zoals het vaste orthopedische ondersteunende OK-team met de orthopedisch staf, samenwerken om de aandacht van de "roulerende" team-leden te richten en vast te houden op deze specifieke biomateriaal-gerelateerde OK attitude.

Een onderzoek om de mogelijke bacteriële contaminatie vast te stellen bij de primaire totale heup arthroplastiek is beschreven in hoofdstuk 3. Veel onderzoek is verricht naar instrumenten die betrokken zijn in het hele operatieve proces. Dit onderzoek richt zich op de instrumenten die in contact zijn geweest met de directe plaats van de te plaatsen prothese. Er zijn bacteriële kweken afgenomen van de raspen gebruikt voor het voorbereiden van het acetabulaire (bekken) en femorale (bovenbeens) bot. Tevens is het op deze wijze verwijderde bot gecontroleerd op bacteriële infectie door middel van kweken, van zowel de acetabulaire als femorale zijde. Dit is gedaan bij 67 opeenvolgende totale heuparthoplastieken van 67 patiënten. De bacterie kweken bij de raspen zijn afgenomen aan het begin en eind van elke specifieke procedure, zowel acetabulair als femoraal. Het totale aantal afgenomen kweken bedroeg 402 waarvan bij 26 een micro-organisme werd gekweekt. Bij 20 patiënten was tenminste 1 van deze positieve kweken afkomstig van materiaal dat in direct contact is geweest met de plaats waar de prothese geplaatst werd. Dit houdt in dat bij tenminste 30% van de betrokken patiënten een positieve bacteriële contaminatie is aangetoond op de plaats van de prothese ten tijde van de operatie.

In hoofdstuk 4 staat de beschrijving en toepassing van de uitgebreide bacterie kweek methode, welke is ontwikkeld door de afdeling Biomedical Engineering van onze universiteit. Deze uitgebreide kweekmethode houdt in dat er langer wordt gekweekt, 7 in plaats van 3 dagen. Tevens wordt het oppervlak van de verwijderde prothese afgeschraapt wat ook 7 dagen wordt gekweekt. Het materiaal is afgenomen bij revisie-operaties van gewrichtsprothesen die los zijn gaan zitten. Als oorzaak van deze loslatingen was bij deze operaties niet de verdenking gerezen op een infectie met micro-organismen. De opzet van deze operaties was dan ook om de zogenaamde a-septische loslatingen in één procedure te reviseren, d.w.z. in één operatie de losgelaten prothese verwijderen en een nieuwe prothese plaatsen. Er zijn 33 protheses of componenten onderzocht. Bij 1 van de 33 gevallen zijn micro-organismen geconstateerd met de standaard bacterie kweekmethode van het ziekenhuis. Bij de uitgebreide kweekmethode groeide er in 14 gevallen een micro-organisme. Bij het kweken van het afgeschraapte materiaal werd nog eens bij 6 extra gevallen een positieve bacterie kweek gevonden. Het totale percentage geïnfecteerde gevallen komt dus neer op 60% (20 van de 33 gevallen). Voor de patiënt kan dit betekenen dat er een nieuwe prothese is geplaatst in geïnfecteerd gebied, waardoor deze patiënt een grote kans loopt dat de prothese opnieuw los gaat laten door de onopgemerkte infectie. Deze resultaten tonen aan dat biomateriaal-gerelateerde infecties onopgemerkt kunnen blijven als er op een standaard wijze gekweekt wordt.

Een onderzoek naar het gebruik van 2-[^{18}F]fluoro-2-deoxy-D-glucose (FDG) bij Positron emission tomography (PET) bij de differentiatie tussen een septische dan wel een aseptische loslating van gewrichtsprothesen is beschreven in hoofdstuk 5. FDG-PET is een onderzoekstechniek waarbij d.m.v. een geringe

hoeveelheid radioactieve tracers een infectieus proces wordt aangetoond.
In deze pilot studie hebben 7 patiënten met een pijnlijke gewrichtsprothese een revisie ingreep ondergaan binnen 6 weken na de PET scan. Uit deze groep hadden 4 patiënten een sterk verhoogde opname van de FDG-tracer. Bij de revisie operatie van deze patiënten werd bij 3 van hen een septische loslating vastgesteld.
Bij de 3 andere patiënten met een pijnlijke gewrichtsprothese werd een milde FDG-activiteit vastgesteld. Bij hen bleek na de revisie operatie sprake te zijn geweest van een a-septische loslating. Bij 5 controle patiënten met 7 asymptomatische gewrichtsprotheses werd een FDG-PET scan verricht vanwege oncologische problematiek. Bij alle PET uitslagen van deze groep patiënten was er in meerder of mindere mate sprake van een vorm van FDG-activiteit. Door de FDG-activiteit bij patiënten met goed functionerende prothesen te bepalen concluderen wij dat het plaatsen van een gewrichtsprothese een mild verhoogde FDG-activiteit laat zien waarschijnlijk als gevolg van het oproepen van een ontstekingsreactie. Een intense FDG-activiteit wordt mogelijk veroorzaakt door reacties op een infectieuze oorsprong.

In hoofdstuk 6 is de preventie van een klinische infectie door middel van gebruik van een 100 µA electrische stroom bij pinnen die percutaan zijn gebracht in het bot van een geit onderzocht. Drie pinnen werden in de rechter achterpoot gebracht bij 9 geiten, waarbij 1 pin voor additionele frame ondersteuning diende.
Twee pinnen werden kunstmatig geïnfecteerd met een *Staphylococcus epidermidis*. Op 1 van de pinnen werd een spanning gezet waardoor een stroompje ging lopen, de andere pen diende als controle. De pingaten werden elke dag gecontroleerd op klinische kenmerken van een infectie. Na 21 dagen werden de geiten opgeofferd en de pinnen uitgenomen.

Het resultaat was dat bij de controlepinnen er bij 89% infecties ontstonden; de andere pinnen vertoonden geen infecties. Deze resultaten tonen aan dat infecties bij percutaan ingebrachte pinnen voorkomen kunnen worden door de toediening van een kleine dosis gelijkstroom.

Het onderwerp van hoofdstuk 7 was de vergelijking van een adhoc benadering met een geprotocolleerde benadering in het geval van persisterende wondlekkage na het inbrengen van een primaire gewrichtsprothese. Een langer lekkende wond na een gewrichtsprothese verhoogt de kans op een geïnfecteerde prothese. Indien deze complicatie vroeg opgemerkt wordt, kan de prothese soms blijven zitten na een schoonmaakoperatie. Lukt dit niet, dan zal de gehele prothese verwijderd moeten worden om de infectie te behandelen.
In dit onderzoek vormden 247 patiënten met 250 prothesen de groep van de adhoc benadering, groep 1. Zij werden behandeld en geobserveerd door een orthopedisch chirurg zonder een duidelijk protocol. 304 patiënten met 308 prothesen vormden groep 2 en deze groep werd geobserveerd en behandeld volgens een geprotocolleerde benadering.
Het percentage van geregistreerde persisterende wondlekkage in groep 2 was bijna 2x groter dan in groep 1 (21% en 11% resp.). Het aantal schoonmaakoperaties was groter in groep 1 (30%) dan in groep 2 (17%). Het percentage prothesen dat gered kon worden door de schoonmaakoperaties was hoger in groep 2 (94%) dan in groep 1 (85%). Het belangrijkste voordeel van een geprotocolleerde benadering werd gezien in de percentages van prothesen die ondanks de persisterende lekkage niet opnieuw geopereerd werden. In groep 2 kon 98% van de prothesen blijven zitten terwijl in groep 1 dat 90% was. Naast een betere registratie maakt de geprotocolleerde benadering het beter mogelijk om die patiënten te selecteren waarbij geen schoonmaakoperatie nodig is.

De algemene discussie beschrijft dat het doen van onderzoek een effectieve manier is om het gedrag van personeel, betrokken in de gehele keten van zorg voor patiënten met een gewrichtsprothese, te optimaliseren. Dit gebeurt door het creëren van een groter bewustzijn en het stimuleren van veranderingen in een breder gebied dan alleen degene die betrokken zijn bij onderzoek (het “Hawthorne-effect”). Dit soort onderzoek wordt van even groot belang verondersteld op lokaal niveau dan de wereldwijde verspreiding van kennis.

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CURRICULUM VITAE

The author of this thesis was born in Enschede, The Netherlands, on September 21th 1963. After graduating from the Jacobus College in 1981 he commenced a study at the Academy of Physical Education. After completing this study he worked as a physical education instructor in the Army. In 1987 he commenced medical school at the University of Groningen. In 1994 he graduated and started to work as a resident at the department of General Surgery of the University Medical Center Groningen.

The first two years of general surgery, as part of the orthopaedic training, was done in Medisch Centrum Leeuwarden. The orthopaedic training was started in the Deventer Ziekenhuis and completed in the University Medical Center Groningen. In 2002 he started to work as an orthopaedic surgeon in the University Medical Center Groningen, with special interest in Pediatric Orthopaedics and Reconstructive Surgery.

He is married to Gaby Maathuis-Sardani and they have a daughter, Tess, and a son, Jesse.

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