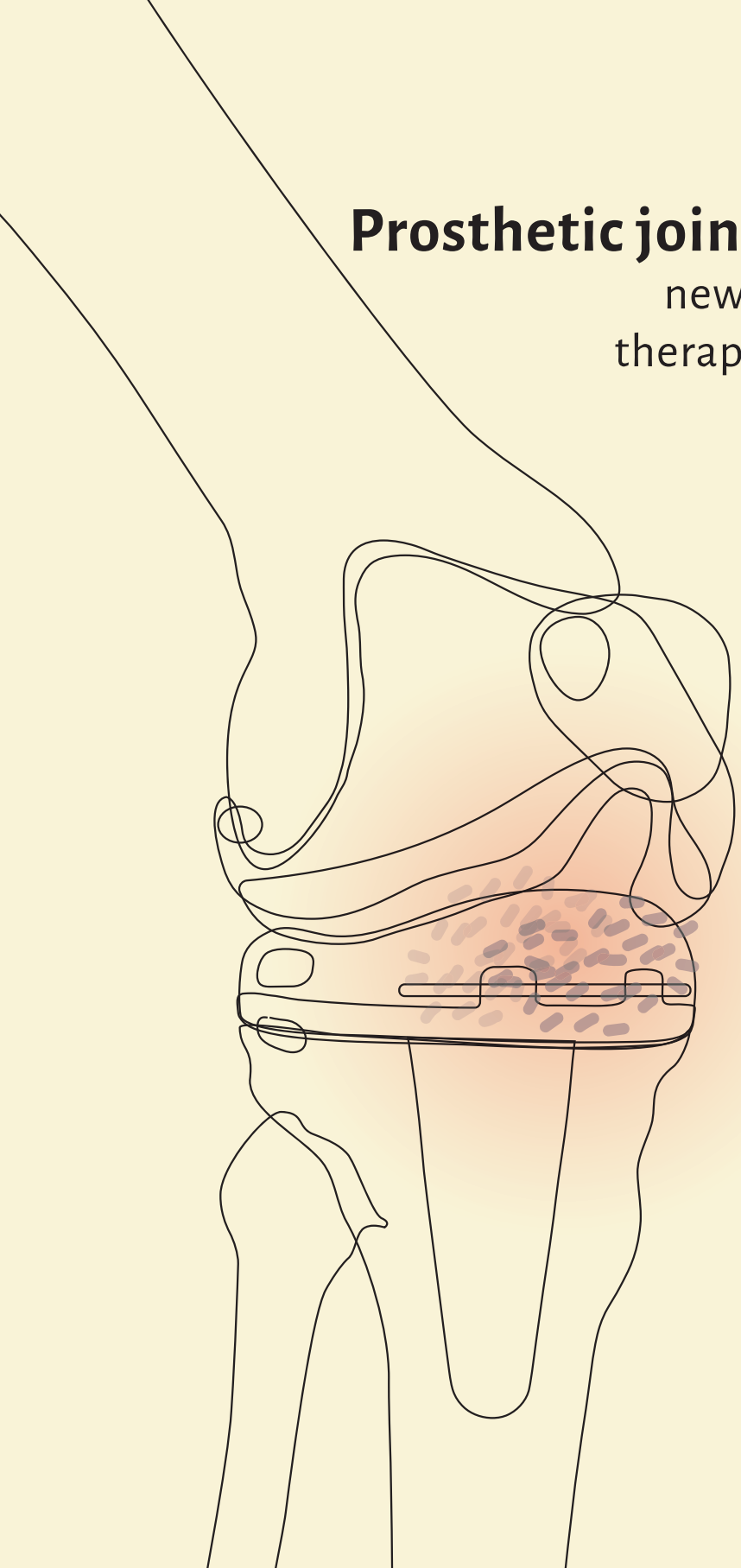
Prosthetic joint infections: new diagnostic and therapeutic strategies

Scheper, H.

Prosthetic joint infections

new diagnostic and
therapeutic strategies



Henk Scheper

Prosthetic joint infections

New diagnostic and therapeutic strategies

Hendrik Scheper

Prosthetic Joint Infections: new diagnostic and therapeutic strategies

ISBN/EAN: 978-94-93315-72-3

Cover & lay-out: Guntra Laivacuma || www.proefschrift-aio.nl
Printed by: Proefschrift all in one || www.proefschrift-aio.nl

Prosthetic joint infections

New diagnostic and therapeutic strategies

Proefschrift

ter verkrijging van
de graad van doctor aan de Universiteit Leiden,
op gezag van de rector magnificus prof. dr. ir. H. Bijl,
volgens besluit van het college voor promoties
te verdedigen op dinsdag 27 juni 2023
klokke 16:15 uur

door

Hendrik Scheper
geboren te Oostflakkee
in 1980

Promotores
Prof. dr. L.G. Visser
Prof. dr. M.G.J. de Boer

Promotiecommissie
Dr. A. Soriano (University of Barcelona)
Prof. dr. F.R. Rosendaal
Prof. dr. R.W. Poolman
Dr. M. Wouthuyzen-Bakker (University Medical Center Groningen)

Table of contents

CHAPTER 1

Introduction and outline of the thesis

Introduction[1]

A prosthetic joint infection (PJI) is a serious complication of arthroplasty often leading to long-term hospitalization, severely restricted mobility and reduced quality of life.[4] The surgical treatment options for PJI are dependent on the chronicity of the infection and host characteristics. In most cases of acute PJI, surgical debridement with retention of the prosthesis, followed by antimicrobial treatment, is the preferred treatment strategy (summarized as 'DAIR': Debridement, Antibiotics and Implant Retention). For chronic PJI, the implant often needs to be removed. Surgical treatment is followed by long-term antimicrobial treatment. Despite well-defined surgical and antimicrobial treatment strategies, failure rates are still considerably high. Reported failure rates vary between 10 and 70% due to heterogeneity in patient populations, type of PJI and different surgical and antimicrobial treatment strategies.[5,6] An important reason for treatment failure is the existence of a biofilm on the surface of the implant. This biofilm consists of a matrix of proteins and nucleic acids in which bacteria can escape the activity of the immune system and can switch to metabolically inactive bacteria, called persisters, against which antibiotics are ineffective.[7] Both the composition and the characteristics of a biofilm explain why curing biofilm-associated infections in general, such as prosthetic joint infections, central line-associated bloodstream infections, infected cardiac devices and vascular graft infections is notoriously difficult. Often, these infections can only be cured if the foreign device is removed, but this is accompanied by major inconvenience for the patient due to prolonged immobility and the need for reoperation to insert a new prosthesis. However, cure may also be achieved after surgical debridement and keeping the prosthesis in situ. It is generally believed that the chance for eradication of infection increases if the time interval between inoculation of bacteria on the implant and surgical debridement is short, although some studies show contradictory results.[8-10] Consequently, a delay in making the diagnosis may cause more chronic infections and lower success rates after DAIR. Removal and delayed reimplantation of the infected joint is an alternative surgical approach for PJI but this is associated with long-term immobility, longer hospital admissions and surgical-related secondary infections. To improve outcome for patients with PJI, both early detection and adequate treatment strategies are important factors for successful treatment. In this introduction, the clinical presentation of acute and chronic PJI and the composition of biofilms are described. Next, a historical overview of the

1 This introduction is based on three earlier publications in NTvG, NVMM and a conference paper from the Boerhaave Posteducational Course on Infectious Diseases in 2018.1. Scheper H, Wouthuyzen-Bakker M, Veldkamp KE, et al. [Prosthetic joint infection]. *Ned Tijdschr Geneeskd* 2019;163, 2. Scheper H RM, M.G.J. de Boer. Geinfecteerde gewrichtsprotheses. Nascholingscursus Infectieziekten 2018. Noordwijkerhout, 2018:27-44, 3. H. Scheper S.A.V van Asten, R.J.P van der Wal, M.G.J. de Boer. Rifampicin for orthopedic infections: a hstorical overview. *Ned Tijdsch Med Microbiol* 2020;28(3):110-17.

antimicrobial treatment strategies for PJI including the use of rifampicin for staphylococcal PJI will be provided. Finally, we formulate the questions underlying this thesis.

In the current literature, PJI is mostly spelled out as prosthetic joint infection but also as periprosthetic joint infection or sometimes prosthetic joint-associated infection. The difference between these terms is merely semantic. A PJI also implicates that bone and surrounding tissues are involved in the infection. In line with the nomenclature that is already in vogue for other biofilm-associated infections, for this thesis the term prosthetic joint infection is consistently used (like in prosthetic valve endocarditis and vascular graft infection).

Epidemiology of PJI

Annually, over 70.000 hip and knee prostheses are implanted in patients in the Netherlands.[11] These operations are generally successful and cost-effective.[12] Due to the aging of the population, the number of implanted joint prostheses, and thus the number of PJI will continue to increase. For the United States of America, it is estimated that more than 3.400.000 prosthetic joints will be implanted in 2030 with a yearly count of 26.000 hip PJIs and 40.000 knee PJIs.[13]

The incidence of PJI after primary arthroplasty is estimated to be 1,5-2% per year.[13,14] After joint revision surgery (approximately 5000 revision procedures per year in the Netherlands), the proportion of PJI is much higher. In the Dutch Arthroplasty Register (LROI), nearly all implanted prosthetic joints are registered. However, in this registry, revision surgery is only recorded if a part of the prosthetic joint is removed or exchanged. Also, patients with PJI who are not operated but only treated with antibiotics are not recorded in this register. Therefore, the actual incidence of PJI is likely to be higher, around 3-5% as recently illustrated in papers from different countries.[15,16]

Pathogenesis and microbiology

Curation of PJI is notoriously difficult due to the formation of a biofilm on the surface of the implant. Known microorganisms associated with biofilm formation are *Staphylococcus aureus*, coagulase-negative staphylococcus (CNS), *Candida albicans*, *Pseudomonas aeruginosa* and *Cutibacterium acnes*, but almost every bacterium can form a biofilm[17]. A biofilm gives a bacterium survival advantage and develops as soon as bacteria adhere to foreign material. The degree of adhesion is determined by, among other things, the nature and roughness of the surface, electromechanical forces, flow velocity around the material, pH, presence of antibiotics, granulocytes, and properties of the bacterium itself (hydrophobicity cell surface, fimbriae, flagellae)[18]. A matrix of extracellular polymeric substances (EPS)

containing proteins, polysaccharides and nucleic acids is formed in which bacteria can proliferate, communicate and escape the activity of antibiotics and the hosts immune system[19]. Most likely, the ineffectiveness of antibiotics to eradicate bacteria from biofilms is not explained by reduced penetration in the biofilm or by antimicrobial resistance. Antibiotics generally penetrate well into biofilms, although sometimes with a delay[20-22]. The main problem of biofilm-associated infections is the presence of persisters[7]. A small proportion of bacteria in the biofilm will, secondary to stress, absence of nutrients or by stochastic variation, switch phenotypically from planktonic bacteria to dormant 'persisters'. Persisters are metabolically inactive bacteria that survive high local antibiotics concentrations because antibiotics can only target dividing bacteria[23]. The knowledge of the existence of persisters dates back to shortly after the introduction of penicillin[24]. In vitro, penicillin was found not to be able to completely kill a population of *S. aureus*. When the small subpopulation of surviving bacteria was incubated again after discontinuing penicillin, it was found to be equally sensitive for penicillin after re-exposure to penicillin. This could be repeated several time without resistance occurring. The surviving bacteria after antibiotic exposure were named persisters.

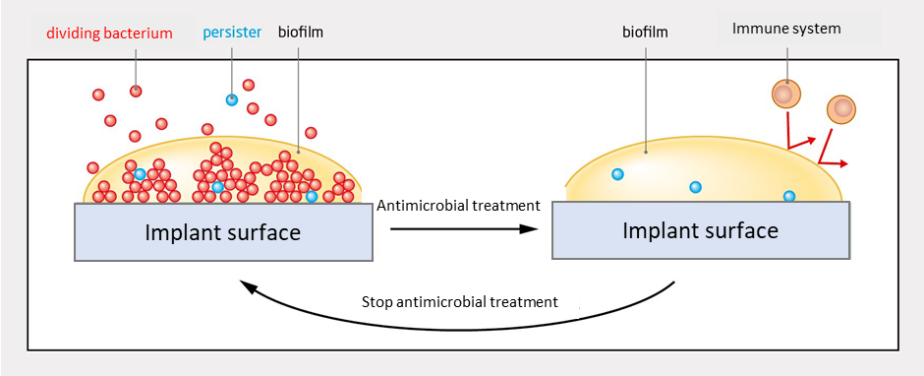


Figure 1. Effect of antimicrobial treatment on bacteria within a biofilm on an implant

In the following decades, limited research has been performed on persisters. During this period, bacteria were referred to as planktonic, free-living, dividing single cells. Since the late seventies, the research landscape changed. The term biofilm was coined in 1981 by Costerton et al. who described the presence of surface-adhering bacteria embedded in a 'glycocalyx' matrix.[25] In the decades thereafter, the molecular mechanisms enabling bacteria to switch to and from a metabolically inactive state were increasingly unraveled.[26-30] The biofilm protects bacteria against eradication by antibiotics and /or the hosts immune defense. Some antibiotics such as rifampicin and fluoroquinolones are able to effectively penetrate a biofilm and therefore often called 'biofilm-active' agents. However, given the dormant state of persisters, it is unlikely that these antibiotics

can eradicate persisters residing in biofilms.[16]. Additionally, it is unknown how long persisters within chronic biofilms are able to survive under chronic antibiotic pressure.

Clinical presentation
The clinical presentation of an acute PJI differs from a chronic PJI. This is because low-virulent pathogens, such as *Staphylococcus epidermidis* and *Cutibacterium acnes*, particularly involved in chronic PJI, lead to a different clinical presentation than, for example, *S. aureus* and streptococci, which due to their virulence give a more acute clinical presentation. Acute PJI during the first weeks after joint arthroplasty can be characterized by acute wound deterioration, swelling of the wound, wound leakage, fever or elevated inflammatory parameters (Table 1)[1]. Acute PJI may also occur years later if a virulent micro-organisms adhere to the prosthesis via haematogenic route or per continuitatem via a nearby focus.

Chronic PJI is characterized by a more prolonged postoperative course of swelling, persistent wound leakage and fistula formation. This may be accompanied by subfebrile body temperature and mildly increased CRP or BSE but inflammatory parameters can also be completely within the normal range. Sometimes, the only clinical sign for a chronic PJI is a loosened prosthesis, or chronic discomfort and/or pain. Differentiation between acute and chronic PJI is relevant because the different causative micro-organisms in acute and chronic PJI require a different antibiotic and surgical strategy. Further, chances for cure appear to decline once an infection becomes more chronic. For practical reasons and to be able to compare studies a schedule of the different clinical presentation was constructed that may be helpful when discussing patients or when comparing patient populations in different studies (Table 1).

Postoperative wound leakage as diagnostic factor for PJI

Persistent postoperative wound leakage is regarded as an important risk factor for PJI.[33-36] However, wound leakage may also be secondary to physiological processes such as fatty necrosis or temporary serosanguinous leakage caused by intraoperative disruption of capillaries.[36] Insight in postoperative wound leakage data is crucial for clinicians who must weigh whether persistent wound leakage in postoperative patients are signs of a PJI, requiring reoperation, or belong to an uncomplicated course, not requiring reoperation. It has been hypothesized that an early DAIR for patients with prolonged wound leakage after arthroplasty may reduce later revision surgery. This is in line with a strong recommendation from an international PJI consensus meeting.[37]

However, quantitative data about postoperative wound leakage after arthroplasty in patients with and without PJI are lacking.[38] This lack of evidence results in a large variety of expert- (but not evidence-) based diagnostic and treatment strategies in daily practice.

Tabel 1. Characteristics of acute and chronic prosthetic joint infection

Characteristic	acute PJI (symptoms ≤ 3 weeks)		chronic PJI (symptoms > 3 weeks)	
	early	**late**	**early**	**late**
Estimated prevalence[31 32]	25%	30%	20%	25%
Time arthroplasty to infection	≤ 3 weeks	months-years	3 weeks - 3 months	> 3 months
Route of infection	exogeneous*	hematogeneous spread or nearby focus	exogeneous*	exogeneous*
Clinical presentation	wound dehiscence, warmth, wound leakage, fever, increase in CRP	acute pain, swelling with or without fever	swelling, warmth, persistent wound leakage, sinus tract, subfebrile temperature, mildly elevated CRP	chronic pain, loosening of prosthesis, sinus tract
Most common causative micro-organisms	*Staphylococcus aureus*, *Enterobacterales*	streptococci, *S. aureus*, *Enterobacterales*	Coagulase-negative staphylococci, enterococci	Coagulase-negative staphylococci, *Corynebacterium Cutibacterium acnes*
Surgical treatment	DAIR	DAIR	DAIR or replacement of prosthesis	replacement of prosthesis

DAIR = 'debridement, antibiotics and implant retention'.

* Infectie due to per- or postoperative colonization of wound.

The likelihood of having a PJI increases if a postoperative patient not only has wound leakage but also other classic signs of infection like fever and redness. Differentiation between a 'superficial' wound infection without involvement of the implant and PJI is extremely difficult. Animal experiments show that in a postoperative wound after arthroplasty only 50-100 bacteria are needed to cause a PJI compared to 10.000-100.000 bacteria needed to cause a wound infection in a postoperative wound without an implant.[39] Therefore, given the high bacterial load in a clinically visible wound infection, it is generally believed that a PJI should be excluded in all patients who present with a wound infection after arthroplasty.. Empirical treatment with antibiotics prior to adequate diagnostics is strongly discouraged, because this delays the diagnostic process, it may lead to false-negative cultures and it will not cure an established PJI after all.

Causative micro-organisms of PJI

The micro-organisms most frequently isolated in patients with PJI are dependent on the type of joint, time elapsed since the implantation and the type of surgery (primary

arthroplasty or tumour reconstruction surgery). For acute knee or hip PJI during the early postoperative period after arthroplasty (usually within the first one month) *Staphylococcus aureus* (30-50%), Coagulase negative staphylococcus (13-30%), *Enterobacteriaceae* (4-16%) and streptococci are the most frequently isolated pathogens 8-16%). Chronic PJI is mainly caused by Coagulase-negative staphylococcus (27-35%), *Cutibacterium acnes* (formerly known as *Propionibacterium acnes*; 6-12%) and polymicrobial flora (911%).[31 32 40] Acute hematogenic infections are predominantly caused by streptococci and *S. aureus* and nearly always monomicrobial.[32 41] In 10-35% of patients, cultures remain negative, although very low culture-negative PJI (1%) was reported in one study.[31 32]. The risk of a negative cultures can be reduced by a standardized method of processing intraoperative cultures and by an antibiotic-free interval before cultures are collected (ideally two weeks prior to diagnosis if possible).

Diagnostic Criteria of PJI

A PJI is suspected based on the clinical presentation, often increased inflammatory parameters and can be confirmed with positive cultures of synovial fluid or intraoperative biopsies or positive histopathological examination. There is not a serological marker with a 100% sensitivity or specificity for PJI. In recent years, a number of organizations have established diagnostic criteria for an prosthetic joint infection (Table 2). The presence of pus, fistula or an identical micro-organism in at least 2 deep-tissue cultures is considered to be definitive evidence for a PJI. Supporting criteria include increased inflammatory parameters, only one positive culture in several deep tissue samples, an increased leukocyte number or an increased percentage of neutrophile granulocytes in the synovial fluid. The release of bacteria from biofilms by sonification appears to be a useful additional tool to increase the sensitivity of cultures, especially in patients with chronic infection with low-virulent microorganisms or in patients pretreated with antibiotics.[42] More research is needed to assess how novel diagnostic biomarkers in synovial fluid and blood can be used as a diagnostic criterium.[43] The European diagnostic criteria were recently updated by the European Bone and Joint Infection Society (Table 2).[44]

Surgical treatment

The most important goal of surgical debridement is the removal of all infected tissue including the biofilm on the prosthesis. In patients with acute PJI, cure of infection with retention of the implant is pursued. In order to achieve this, extensive surgical debridement is needed, also called DAIR (Debridement, Antibiotics and Implant Retention, see Figure 2). The surgically accessible parts of the implant are thoroughly cleaned and the replaceable prosthesis parts ('polyethylene liner', femoral head, insert ('insert') of the acetabulum) are replaced. For patients with chronic PJI or patients with a loose prosthesis infection, chances to cure PJI are much lower when treated with DAIR as described above.[45] In general,

these patients needs removal of the prosthesis during surgical debridement, followed by antibiotic treatment and delayed reimplantation during a second operation. This is called a two-stage exchange procedure. The reimplantation of a new prosthetic joint can also be performed immediately during the same surgical procedure in which the infected joint is removed, called a one-stage exchange. Observational studies show a reinfection rated between 0% and 41% in two-stage exchange studies and between 0% and 11% in one-stage exchange studies, but no randomized controlled trials have been published yet.[46]

Antimicrobial treatment

The antibiotic therapy is directed against the infection in the tissue around the prosthesis and against surviving bacteria on the implant after surgical debridement. During the postoperative period after debridement, new biofilm formation after debridement needs to be prevented at all costs. Therefore, effective treatment should be started a soon as possible after the operation. The choice of antimicrobial strategy depends on the susceptibility of the causative pathogen, the comedication that is used, documented allergies or intolerances and hosts factors like medical history and patient adherence. Patients are treated with long-term antimicrobial treatment, usually between six and twelve weeks.[47] In the recently published DATIPO trial 6 weeks of antibiotic therapy were inferior to 12 weeks of antibiotic therapy in patients with PJI treated with DAIR or one-stage exchange.[48] This study contradicted the results of a number of observational studies in which a shorter treatment period (6-8 weeks) was also sufficient.[49-51] Failure of treatment may be secondary to residual (infected) cement, a secondary infection with another micro-organism after surgical debridement, improper antibiotic use, reduced compliance to therapy, or antibiotic resistance of the causative micro-organism. Often, in these patients the prosthetic joint needs to be removed.

The use of rifampicin-based antibiotic strategies for staphylococcal PJI

Many studies have reported outcome of PJI after surgical debridement in order to evaluate an antibiotic treatment strategy for staphylococcal PJI. Most of these studies were observational retrospective studies. For staphylococcal PJI, two randomized controlled trials were published about the adjunctive value of rifampicin combination therapy for staphylococcal PJI. Over the last decennia, rifampicin has become the cornerstone of treatment for acute staphylococcal PJI.[52 53] Unfortunately, the use of rifampicin is hampered by drug-drug interactions and significant side effects.[54] Therefore, safe and effective alternative antimicrobial regimens for PJI are needed, but comparative data evaluating a rifampicin-based strategy with other antimicrobial strategies, such as clindamycin, levofloxacin or flucloxacillin, are nearly absent. This paragraph gives a historical overview of the (history of the) role of rifampicin combination therapy for orthopedic infections.

Table 2. Diagnostic criteria for PJI according to American and European scientific societies

	MSIS 2011	IDSA 2013	ICM 2018	EBJIS 2021
Confirmative criteria*				
pus around the prosthetic joint		•		
sinus tract	•	•	•	•
same microorganism in ≥ 2 intraoperative samples	•	•	•	•
virulent microorganism in ≥ 1 intraoperative sample		•		
leukocyte count >3000/µl or >80% PMN in synovial fluid				•
Positive α-defensin in synovial fluid				•
acute inflammation in histopathologic examination‡				•
>50CFU/ml any organism on sonication				•
Supportive criteria‖				
Preoperative:				
Clinical features: early radiographic loosening, CRP>10, wound healing problem, purulence around prosthesis, recent fever or bacteremia				•
elevated CRP- of D-dimer concentration	•		• (2 p)	
ESR > 30 mm/h			• (1 p)	
Positive α-defensin in synovial fluid			• (3 p)	
CRP > 6,9 mg/l in synovial fluid			• (1 p)	
leukocyte count > 3000/µl in synovial fluid	•		• (3 p)	
leukocyte count > 1500/µl or >65% PMN in synovial fluid				•
> 80% granulocytes in synovial fluid			• (2 p)	•
Intraoperative:				
pus around the prosthetic joint	•		• (3 p)	•
virulent microorganism in ≥ 1 intraoperative sample	•	•	• (2 p)	•
acute inflammation in histopathologic sample ‡	•	•	• (3 p)	
>1 CFU/ml any organism on sonication				•

IDSA = Infectious Diseases Society of America; MSIS = Musculoskeletal Infection Society; ICM = International Consensus Meeting on Musculoskeletal Infection (Philadelphia, 2018); EBJIS = European Bone and Joint Infection Society. ESR = erythrocyte sedimentation rate, CRP = C-reactive protein. PMN = polymorphonuclear neutrophils. P = points.

* PJI is confirmed, according to this societies in the presence of >1 confirmative criteria

‡ Presence of ≥ 5 neutrophils in ≥ 5 high-powerfields (400 x) *or* visible micro-organisms

‖ According to MSIS, PJI is confirmed in the presence of at least 4 supporting criteria. According to ICM, *preoperatively*, PJI is confirmed if at least 6 p, PJI is likely if 2 to 5 p; no PJI if 0 to 1 point. *Intraaoperatively*, PJI is confirmed if at least 6 p; PJI may be present if 4 to 5 p, no PJI if 0 to 3 p. According to EBJIS, PJI confirmed if at least 1 confirmative criterium, PJI likely if combination of 2 supporting criteria, requiring 1 clinical feature and 1 laboratory feature

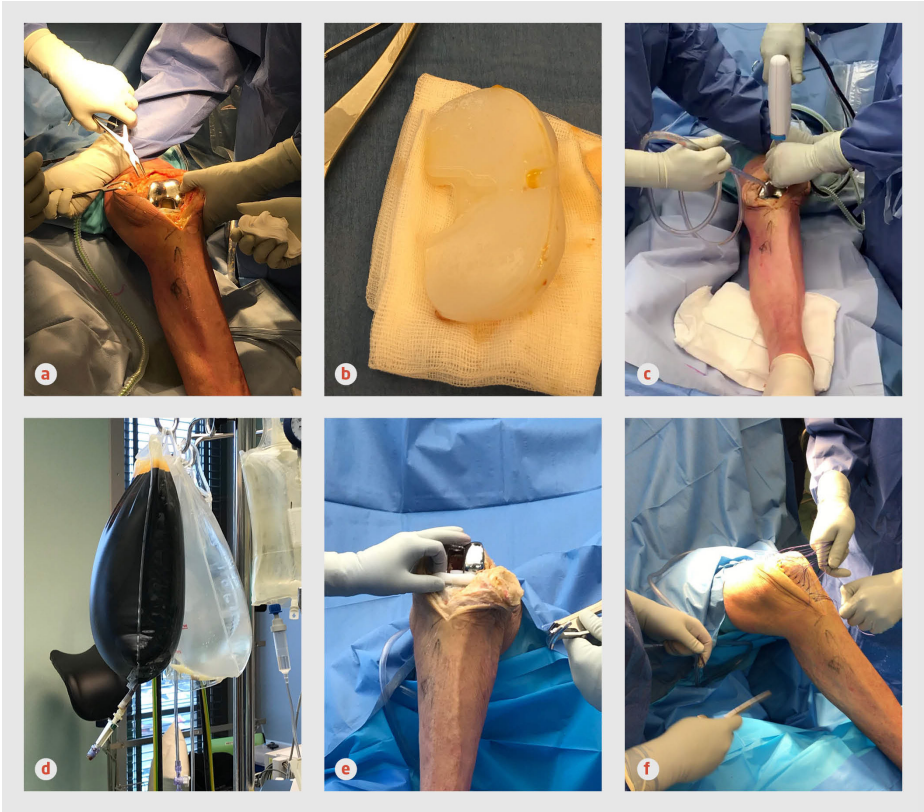


Figure 2. Surgical debridement of an infected knee implant

Legend. (a) During debridement, the joint is opened, multiple deep-tissue cultures are taken and infected tissue is removed. (b) The exchangeable prosthetic parts, such as this liner of the knee prosthesis, are replaced. After removing the liner, the surgeon also has better access to the surgical area that needs to be de debrided. (c) and (d). The joint is rinsed extensively with sodium chloride mixed with an antiseptic such as iodine with the 'pulsed lavage' technique. (e) Then, the surgical area is re-covered with clean drapings and a new liner is inserted. (f) Afterwards the wound is closed. (Scheper et al. Ned Tijdschrift v Geneeskunde 2019, *pictures obtained from Drs. R Mahdad, Alrijne hospital*)

The discovery of rifampicin

Rifampicin owes its name to Piero Sensi, an Italian scientist who had the habit of giving nicknames to newly discovered antibiotics. In 1957, his Milanese research group isolated, from a French soil sample, a new class of antibiotics from the bacterium *Amycolatopsis rifamycinica* (previously called *Streptomyces mediterranei* and subsequently called *Nocardia mediterranea*).[55] This new antibiotic class, called rifamycins, were named after the then famous French gangster film Rififi (French for "trouble"), directed by Jules Dassin. The abbreviation of the active ingredient (N-**A**mino-N-**M**ethyl**P**iperazine) completed the name. The first rifamycin for clinical use, rifamycin SV, was replaced by rifampicin due

to better bioavailability and effectiveness, especially against *Mycobacterium tuberculosis*. Later it was also found to be very effective against *Mycobacterium leprae*.[56] Rifampicin kills bacteria by binding to the β-subunit of the DNA-dependent RNA polymerase, resulting in blocking of the transcription process and consequent inhibition of bacterial RNA synthesis.[57] Rifampicin diffuses freely into tissues and bacteria and is highly effective against Gram-positive and some Gram-negative bacteria and also has bactericidal activity on intracellular microorganisms. The drug came on the market in 1968, and is now, more than 50 years later, still an essential part of tuberculostatic treatment. Resistance against rifampicin quickly occurs by a single mutation in the rpoB gene, which encodes the β-subunit of the RNA polymerase.[57] This resistance may occur within 48 hours when rifampicin is used as monotherapy or in the treatment of infections with a very high bacterial load[58]. The fear for widespread rifampicin resistance in such a powerful anti-tuberculous drug resulted in a strong lobby, led by pulmonologists, to discourage the use of rifampicin for non-tuberculous infections. However, that fear turned out to be unjustified in 1980 when a study showed that the incidence of rifampicin-resistant tuberculosis strains in countries where rifampicin was freely used in combination therapy for urinary and respiratory infections was not higher than in countries with highly restrictive use of rifampicin only for tuberculosis.[59]

Experimental animal studies about PJI

In 1973, Mandell et al. reported that of 18 antibiotics tested, only rifampicin was able to kill intracellularly dividing bacteria in macrophages that had survived phagocytosis.[60] Interestingly, rifampicin did not kill inactive intracellular bacteria that had been metabolized by cooling to non-dividing state. This led the authors to suspect that there was no specific mechanism of action for rifampicin, but mainly a good intracellular penetration. Later, several studies confirmed the excellent penetration of rifampicin into biofilms, but persisters could not be killed with rifampicin.[61 62] In 1983, the group of Waldvogel showed in the first animal study that rifampicin monotherapy, when administered three hours before to twelve hours after inoculation of *S. aureus* in a Teflon cage which was placed in the flank of guinea pigs, prevented infection[63]. If rifampicin was administered after twelve hours of inoculation, infection developed with growth of rifampicin-resistant *S. aureus*. Since then, at least seven experimental animal studies in guinea pigs with implanted Teflon cages have been performed by groups directed by Zimmerli et al.[64] In these studies, the time from inoculation to initiation of antibiotics was between 24 and 72 hours. The duration of treatment with rifampicin lasted four days. Betalactam antibiotics and clindamycin could not be tested because these antibiotics induced severe weight loss in the guinea pigs within four days. The cure rate with rifampicin/fluoroquinolone combination therapy in the guinea pigs in these studies was 88-100%. Monotherapy with fluoroquinolones, vancomycin, daptomycin or

linezolid resulted in a cure of 0% in six out of seven studies. In another experimental animal model, rats with artificial implants were treated, after 14 days of inoculation with MRSA, with six days of rifampicin combination therapy. None of the animals were cured.[65]

In all the described animal experiments above, debridement of the infected implant was not part of treatment. Apparently, the duration of biofilm formation and possibly also the innate immune system of the type of animal have a crucial influence on the effectiveness of the antibiotic treatment.

Rifampicin for PJI: outcome in randomized controlled trials

In 1974 Bourret et al. in the *Lyon Medical* were the first to report a 'very satisfactory clinical effect' with rifampicin combination therapy for osteomyelitis caused by staphylococci (original article not traceable anymore). In 1979 some clinical studies were summarized in which the usefulness of rifampicin was also described in other non-tuberculous microorganisms. Rifampicin combination therapy seemed particularly promising for staphylococcal infections.[66] In a small randomized prospective study in the 1980s, patients with chronic *S. aureus* osteomyelitis (i.e. without artificial material) were randomized between nafcillin monotherapy (cure 4 out of 8 patients) and nafcillin-rifampicin combination therapy (cure 8 out of 10 patients).[67] Next, a randomized controlled trial (RCT) in 101 patients with *S. aureus* infection showed no benefit with rifampicin combination therapy; in a subgroup of 23 osteomyelitis patients, rifampicin/oxacillin (or rifampicin/vancomycin) combination therapy appeared to be more effective.[68] In 1992, for the first time, a small cohort was published describing cure in in 11 patients with an orthopedic infection (82%).[69] This study prompted a randomized controlled trial, led by Zimmerli et al., which was published in 1998 and became the most cited article on PJI, reaching >1500 citations.[52] In this study, consisting of 18 patients with PJI and 15 patients with osteosynthesis-associated infection, the effect of ciprofloxacin monotherapy compared to ciprofloxacin with rifampicin was evaluated. Patients were randomized between (a) two weeks of beta-lactam antibiotics followed by ciprofloxacin monotherapy and (b) two weeks of beta-lactam antibiotics with rifampicin (2dd450mg) followed by ciprofloxacin with rifampicin. The rifampicin in the treatment group was started immediately postoperatively. Duration of treatment was 3 months (for hip PJI and osteosynthesis-associated infection) or 6 months (for knee PJI). Dropout occurred in 6 of 18 patients in the rifampicin group (33%). Curation in the rifampicin group was 89%, in the placebo group 60% (intention-to-treat analysis $p=0.10$, per protocol analysis $p<0.02$). This difference in outcome was mostly explained by the development of ciprofloxacin-resistant staphylococci in 4 out of 5 failures in the ciprofloxacin monotherapy group. Unfortunately, the outcome was not stratified for type of infection(PJI versus

osteosynthesis-associated infection) or causative agent (CNS versus *S. aureus*). Also, the study was heavily underpowered although the beneficial effect of rifampicin combination therapy was striking and distinctly different with a few small cohort studies from the 1990s. In those cohort studies the success rate after DAIR for staphylococcal PJI did not exceed 40%.[70 71] In 2020, a second randomized controlled trial was published in which 48 patients with a staphylococcal PJI were randomized between rifampicin combination therapy and beta-lactam monotherapy.[72] Treatment success was comparable in both groups: 72% and 74%. This study was also underpowered, although the study contained three times as much patients as the trial by Zimmerli et al.

Observational cohort studies about PJI:

In many observational studies the role of rifampicin for staphylococcal prosthetic joint infection was evaluated. Since 2005, more than 60 observational studies have been published describing the outcomes of DAIR in staphylococcal prosthetic joint infections, of which less than 30 studies also evaluated the role of rifampicin. Numerous studies, comparing rifampicin combination therapy to a treatment strategy without rifampicin, reported no positive association between rifampicin and treatment success [49 73-79], while other studies showed a positive correlation.[5 80 81] With rifampicin use, success rates are 50-90%, while in patients without rifampicin (described in about ten studies) it varies between 30 and 80%. The cure rate for infected hip prostheses in these studies was in some but not all studies higher than for infected knee prostheses[82]. In addition, in studies in which the use of rifampicin in staphylococcal infections was not described, the cure rate was lower than in the studies in which rifampicin was given.[82] Reported success rates gradually increase over the years, but remain at around 50% in several large cohorts with more than 150 enrolled patients with staphylococcal infections.[83-87] Differences in outcome may be explained by differences in proportion of included polymicrobial PJI, differences in surgical approach, like whether or not mobile parts were exchanged changed during debridement. Different definitions of treatment success are also a relevant factor. The literature regarding the role of rifampicin for staphylococcal PJI has not been systematically appraised yet. The effectivity of antimicrobial monotherapy without rifampicin for staphylococcal or streptococcal PJI has not been evaluated in PJI either. Some observational studies have reported about monotherapy with moxifloxacin, flucloxacillin, or linezolid for staphylococcal PJI, showing reasonable outcomes.[72 73 88 89]

Methodological limitations of observational studies

Selection bias, confounding by indication and survival bias are important methodological limitations of the observational studies discussed here. Selection bias occurs because, due to toxicity and interactions, rifampicin may be less prescribed in vulnerable patients who already have an a priori increased chance of a worse outcome. In addition,

only patients with rifampicin sensitive *S. aureus* are selected for rifampicin treatment. Immortal time bias occurs when rifampicin is withhold in patients until one to two weeks after debridement (after the wound is dry and the sensitivity of the staphylococcus is known). Only patients who do not develop a failure during those two weeks will start with rifampicin, leading to better outcomes in the rifampicin group.[81 90] Confounding by indication can occur because, for example, physicians who prescribe rifampicin are better aware of current guidelines and may also be more skilled surgeons performing who may exchange mobile parts more often during surgery. In most observational studies, the patient characteristics of the groups with and without rifampicin combination therapy were not reported, making correction for these confounders challenging.

Timing and duration of use of rifampicin

To prevent new biofilm formation after debridement by surviving bacteria that attach to the retained implant, rifampicin-combination therapy should be started as soon as possible after surgical debridement. The highly bactericidal activity of rifampicin will rapidly kill any remaining bacteria in the postoperative wound. If surviving bacteria in the postoperative wound attach to the prosthetic joint and cause new biofilm formation, the risk of treatment failure will be high. Therefore, in the very early postoperative period, rifampicin may be mostly needed. Most clinicians however withhold postoperative rifampicin treatment until the wound is dry and the rifampicin sensitivity of the staphylococcus is known. This is due to the presumed risk of selecting for rifampicin-resistant Coagulase-negative skin staphylococci that may secondary infect the implant through the postoperative leaking wound. Although this view is widely accepted, it is not supported by clinical studies that demonstrate relapsing PJI by rifampicin-resistant staphylococcal when administered immediately postoperative. In vitro research shows that rifampicin resistance only develops with in the presence of a high bacterial load and if rifampicin is used as monotherapy. In a patient with a PJI, the bacterial load is significantly reduced during the surgical debridement and patients are always treated with rifampicin combination therapy. In the earlier mentioned RCT of Zimmerli et al. rifampicin was started immediately postoperative, not resulting in rifampicin-resistant staphylococci in patients who failed on therapy[52]. Therefore, it may be justified not to withhold this excellent anti-staphylococcal drug in the early postoperative period in which it may be mostly needed. More data to support this strategy are needed.

The role of chronic suppressive antibiotic therapy

Chronic suppressive antibiotic treatment for PJI is the chronic use of low-dose antibiotics in patients with a (relapsing) PJI who are no longer eligible for surgery or who decide not to undergo further surgery. Chronic suppressive antibiotic therapy is only started after the normal treatment duration for PJI (usually between six and twelve weeks) and is aimed at

f controlling rather than curing the chronic PJI. The rationale behind suppressive therapy is that the persisters within the biofilm that have survived antimicrobial treatment and switch back to a metabolically active state, cannot proliferate further due to a daily rise in antibiotic concentrations. The choice for a certain regimen is dependent on several criteria: (a) the micro-organism is well sensitive to the antibiotic; (b) antibiotics can be taken orally; (c) the safety of long-term use is known; and (d) the antibiotics have a reasonable penetration into bone and joint tissue. The dose of chronic suppressive therapy is usually lower than during conventional antibiotic therapy, because no more tissue infection or osteomyelitis needs to be treated.

Data about the optimal dosing and duration of suppressive antibiotic treatment are absent. Some centers advocate lifelong continuation with suppressive therapy while others state that treatment may be stopped after a two to five years, provided that inflammatory parameters remain low and clinical signs for infection are absent. The treatment strategy and duration for inoperable chronic PJI should always be determined in a multidisciplinary team meeting.

Table 3. Expert-based regimens of antibiotic suppressive therapy used at Leiden University Medical Center

Flucloxacillin 1000mg BD
Doxycyclin 100mg OD
Amoxicillin 1000mg BD
Cotrimoxazol 960mg OD (sometimes lowered further to 480mg OD)
Clindamycin 600mg BD (sometimes lowered further to 300mg BD)

Need for innovative treatment options for PJI

Despite an increasing amount of knowledge about the optimal diagnostic and treatment strategies for PJI, cure rates after treatment remain disappointingly low.[5 6 87]. In several of the largest studies, most closely approximating the real-life clinical situation, cure rates were between 50-70%. This low care rate likely relates to surviving bacteria within a mature biofilm, even after thorough surgical debridement and adequate antimicrobial treatment. Therefore, innovative alternatives for antibiotics, that may act synergistically with antibiotics, are urgently needed. New therapeutic modalities, such as immunotherapy, nanoparticles, bacteriophages, photodynamic therapy, heat induction, novel antibiotics and antimicrobial peptides are several promising complimentary treatment strategies for PJI and need to be tested further.[91 92] In addition, we are in need for in vitro and experimental animal models, that approximate a PJI as much as possible in which the most promising strategies can be further tested in order to enhance future cure rates for PJI.

Outline of the thesis

In the preceding paragraphs, the challenges and knowledge gaps clinicians face when treating patients with PJI have been described. To improve outcome for patients with PJI, high-quality studies are needed to both improve the diagnostic process as well as patient-tailored antimicrobial treatment strategies. As described in this introduction, earlier diagnosis of PJI may enhance success rates. Further, evidence-based antimicrobial treatment strategies are needed for PJI. Lastly, the high rate of relapses after treatment for PJI, even if treated according to the best available level of evidence, urges us to explore novel treatment strategies aimed at eradication of persisters from the biofilm. The current thesis addresses these challenges. The thesis is divided by three parts focused on new diagnostic and antimicrobial strategies.

Part I. The use of E-health to detect prosthetic joint infections

A general introduction to the topic of this thesis is described in **chapter 1**. The *first* part focuses on earlier detection of PJI. A postoperative woundcare app was developed to shorten the time to diagnosis of PJI and to assess the association between postoperative wound leakage and occurrence of PJI. In **chapter 2**, we evaluated the ease of use and perceived usefulness in a group of patients who used the woundcare app in a pilot study. In **chapter 3**, we compared the extent and duration of postoperative wound leakage in patients with and without PJI, using the same smartphone application, in a large multicenter implementation study in The Netherlands.

Part II. Evaluation of current antimicrobial strategies for PJI

The *second* part of this thesis focuses on the evaluation of currently used antimicrobial treatment strategies for staphylococcal PJI. In **chapter 4**, the outcome of PJI in all studies reporting the outcome of staphylococcal PJI after DAIR is assessed in a systematic review and meta-analysis, focused on the use of rifampicin for staphylococcal PJI. In **chapter 4,** we also describe the importance of acknowledging several forms of bias and confounding that are inevitably present in most observational studies about PJI and we discuss how to correctly deal with these. **Chapter 5** specifically focuses on causative micro-organisms and outcome for different surgical strategies for patients with an infected megaprosthesis, a subgroup of patients that is even more challenging to treat compared to PJI after conventional arthroplasty.

Part III. New antimicrobial strategies for PJI

New strategies to combat biofilm-associated infections like PJI are the subject of the *third* part of this thesis. **Chapter 6** describes the results of a large, prospective cohort of

patients with staphylococcal PJI who were treated in our region, according to a predefined protocol, with either a short-term rifampicin strategy or a long-term rifampicin strategy, depending on the hospital patients were admitted to. This large prospective quality registry started after publication of the outcome of staphylococcal PJI treated with DAIR and only short induction therapy with rifampicin is in a small, retrospective observational study ,also described in **chapter 6**. In **chapter 7**, we report on the development of an in vitro biofilm model simulating PJI as much as possible. In this study, the effectivity of several promising anti-biofilm and anti-persister agents was assessed in pretreated mature biofilm models.

Chapter 8 provides a general discussion. The main conclusions of the thesis are highlighted and the implications of our research are put into future perspective.

References

1. Scheper H, Wouthuyzen-Bakker M, Veldkamp KE, et al. [Prosthetic joint infection]. *Ned Tijdschr Geneeskd* 2019;163 [published Online First: 2019/05/24]
2. Scheper H RM, M.G.J. de Boer. Geinfecteerde gewrichtsprotheses. Nascholingscursus Infectieziekten 2018. Noordwijkerhout, 2018:27-44.
3. H. Scheper SAVvA, R.J.P van der Wal, M.G.J. de Boer. Rifampicin for orthopedic infections: a hstorical overview. *Ned Tijdsch Med Microbiol* 2020;28(3):110-17.
4. Sabah SA, Alvand A, Price AJ. Revision knee replacement for prosthetic joint infection: Epidemiology, clinical outcomes and health-economic considerations. *Knee* 2021;28:417-21. doi: 10.1016/j.knee.2020.12.024 [published Online First: 2021/01/28]
5. Lora-Tamayo J, Murillo O, Iribarren JA, et al. A large multicenter study of methicillin-susceptible and methicillin-resistant Staphylococcus aureus prosthetic joint infections managed with implant retention. *Clin Infect Dis* 2013;56(2):182-94. doi: cis746 [pii];10.1093/cid/cis746 [doi]
6. Lora-Tamayo J, Senneville E, Ribera A, et al. The Not-So-Good Prognosis of Streptococcal Periprosthetic Joint Infection Managed by Implant Retention: The Results of a Large Multicenter Study. *Clin Infect Dis* 2017;64(12):1742-52. doi: 10.1093/cid/cix227 [published Online First: 2017/04/04]
7. Lewis K. Persister cells. *Annu Rev Microbiol* 2010;64:357-72. doi: 10.1146/annurev.micro.112408.134306 [doi]
8. Tsang SJ, Ting J, Simpson A, et al. Outcomes following debridement, antibiotics and implant retention in the management of periprosthetic infections of the hip: a review of cohort studies. *Bone Joint J* 2017;99-B(11):1458-66. doi: 10.1302/0301-620X.99B11.BJJ-2017-0088.R1 [published Online First: 2017/11/03]
9. Sendi P, Lotscher PO, Kessler B, et al. Debridement and implant retention in the management of hip periprosthetic joint infection: outcomes following guided and rapid treatment at a single centre. *Bone Joint J* 2017;99-B(3):330-36. doi: 10.1302/0301-620X.99B3.BJJ-2016-0609.R1 [published Online First: 2017/03/03]
10. Ottesen CS, Troelsen A, Sandholdt H, et al. Acceptable Success Rate in Patients With Periprosthetic Knee Joint Infection Treated With Debridement, Antibiotics, and Implant Retention. *J Arthroplasty* 2019;34(2):365-68. doi: 10.1016/j.arth.2018.09.088 [published Online First: 2018/11/08]
11. Jaarraportage Landelijke Registratie Orthopedische Implantaten 2018. *wwwlroi-rapportagenl*
12. Losina E, Walensky RP, Kessler CL, et al. Cost-effectiveness of total knee arthroplasty in the United States: patient risk and hospital volume. *Arch Intern Med* 2009;169(12):1113-21; discussion 21-2. doi: 10.1001/archinternmed.2009.136 [published Online First: 2009/06/24]
13. Premkumar A, Kolin DA, Farley KX, et al. Projected Economic Burden of Periprosthetic Joint Infection of the Hip and Knee in the United States. *J Arthroplasty* 2021;36(5):1484-89 e3. doi: 10.1016/j.arth.2020.12.005 [published Online First: 2021/01/11]
14. Kim HS, Park JW, Moon SY, et al. Current and Future Burden of Periprosthetic Joint Infection from National Claim Database. *J Korean Med Sci* 2020;35(49):e410. doi: 10.3346/jkms.2020.35.e410 [published Online First: 2020/12/23]
15. Kamp MC, Liu WY, Goosen JHM, et al. Mismatch in Capture of Periprosthetic Joint Infections Between the Dutch Arthroplasty Register (LROI) and a Detailed Regional Periprosthetic Joint Infection Registry. *J Arthroplasty* 2021 doi: 10.1016/j.arth.2021.09.001 [published Online First: 2021/09/27]

16. Sinagra ZP, Davis JS, Lorimer M, et al. The accuracy of reporting of periprosthetic joint infection to the Australian Orthopaedic Association National Joint Replacement Registry. *Bone Jt Open* 2022;3(5):367-73. doi: 10.1302/2633-1462.35.BJO-2022-0011.R1 [published Online First: 2022/05/06]
17. Tande AJ, Patel R. Prosthetic joint infection. *Clin Microbiol Rev* 2014;27(2):302-45. doi: 10.1128/CMR.00111-13 [published Online First: 2014/04/04]
18. Busscher HJ, van der Mei HC. How do bacteria know they are on a surface and regulate their response to an adhering state? *PLoS pathogens* 2012;8(1):e1002440. doi: 10.1371/journal.ppat.1002440 [published Online First: 2012/02/01]
19. Koo H, Allan RN, Howlin RP, et al. Targeting microbial biofilms: current and prospective therapeutic strategies. *Nature reviews Microbiology* 2017;15(12):740-55. doi: 10.1038/nrmicro.2017.99 [published Online First: 2017/09/26]
20. Dunne WM, Jr., Mason EO, Jr., Kaplan SL. Diffusion of rifampin and vancomycin through a Staphylococcus epidermidis biofilm. *Antimicrob Agents Chemother* 1993;37(12):2522-6. [published Online First: 1993/12/01]
21. Stewart PS. Antimicrobial Tolerance in Biofilms. *Microbiology spectrum* 2015;3(3):10.1128/microbiolspec.MB-0010-2014. doi: 10.1128/microbiolspec.MB-0010-2014
22. Jefferson KK, Goldmann DA, Pier GB. Use of confocal microscopy to analyze the rate of vancomycin penetration through Staphylococcus aureus biofilms. *Antimicrob Agents Chemother* 2005;49(6):2467-73. doi: 10.1128/aac.49.6.2467-2473.2005 [published Online First: 2005/05/27]
23. Lewis K. Persister cells, dormancy and infectious disease. *Nat Rev Microbiol* 2007;5(1):48-56. doi: nrmicro1557 [pii];10.1038/nrmicro1557 [doi]
24. Bigger J. Treatment of staphylococcal infections with penicillin by intermittent sterilisation. *The Lancet*;244(6320):497-500. doi: 10.1016/S0140-6736(00)74210-3
25. Costerton JW, Stewart PS, Greenberg EP. Bacterial biofilms: a common cause of persistent infections. *Science* 1999;284(5418):1318-22. doi: 10.1126/science.284.5418.1318 [published Online First: 1999/05/21]
26. Conlon BP, Rowe SE, Lewis K. Persister cells in biofilm associated infections. *Advances in experimental medicine and biology* 2015;831:1-9. doi: 10.1007/978-3-319-09782-4_1 [published Online First: 2014/11/12]
27. Conlon BP, Nakayasu ES, Fleck LE, et al. Activated ClpP kills persisters and eradicates a chronic biofilm infection. *Nature* 2013;503(7476):365-70. doi: nature12790 [pii];10.1038/nature12790 [doi]
28. Shan Y, Brown Gandt A, Rowe SE, et al. ATP-Dependent Persister Formation in Escherichia coli. *mBio* 2017;8(1) doi: 10.1128/mBio.02267-16 [published Online First: 2017/02/09]
29. Balaban NQ, Merrin J, Chait R, et al. Bacterial persistence as a phenotypic switch. *Science* 2004;305(5690):1622-5. doi: 10.1126/science.1099390 [published Online First: 2004/08/17]
30. Lewis K. Persister cells and the riddle of biofilm survival. *Biochemistry (Mosc)* 2005;70(2):267-74. doi: BCM70020327 [pii]
31. Zeller V, Kerroumi Y, Meyssonnier V, et al. Analysis of postoperative and hematogenous prosthetic joint-infection microbiological patterns in a large cohort. *The Journal of infection* 2018 doi: 10.1016/j.jinf.2017.12.016 [published Online First: 2018/02/06]

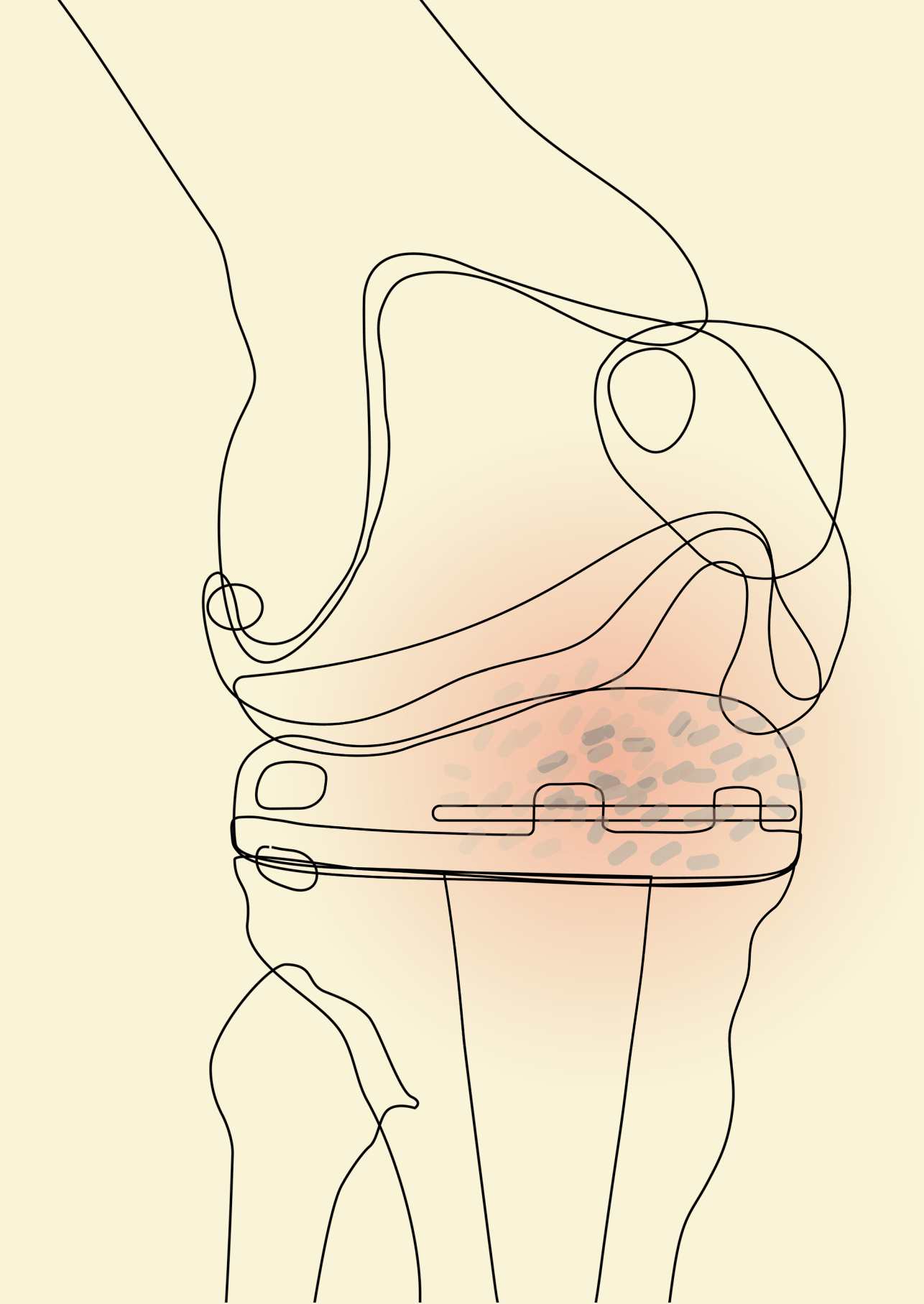
32. Manning L, Metcalf S, Clark B, et al. Clinical Characteristics, Etiology, and Initial Management Strategy of Newly Diagnosed Periprosthetic Joint Infection: A Multicenter, Prospective Observational Cohort Study of 783 Patients. *Open Forum Infect Dis* 2020;7(5):ofaa068. doi: 10.1093/ofid/ofaa068 [published Online First: 2020/05/21]
33. Almeida RP, Mokete L, Sikhauli N, et al. The draining surgical wound post total hip and knee arthroplasty: what are my options? A narrative review. *EFORT Open Rev* 2021;6(10):872-80. doi: 10.1302/2058-5241.6.200054 [published Online First: 2021/11/12]
34. Patel VP, Walsh M, Sehgal B, et al. Factors associated with prolonged wound drainage after primary total hip and knee arthroplasty. *J Bone Joint Surg Am* 2007;89(1):33-8. doi: 10.2106/JBJS.F.00163 [published Online First: 2007/01/04]
35. Wagenaar FBM, Lowik CAM, Zahar A, et al. Persistent Wound Drainage After Total Joint Arthroplasty: A Narrative Review. *J Arthroplasty* 2019;34(1):175-82. doi: 10.1016/j.arth.2018.08.034 [published Online First: 2018/09/25]
36. Kurtz SM, Lau E, Watson H, et al. Economic burden of periprosthetic joint infection in the United States. *J Arthroplasty* 2012;27(8 Suppl):61-5 e1. doi: 10.1016/j.arth.2012.02.022 [published Online First: 2012/05/05]
37. Parvizi J, Gehrke T, Chen AF. Proceedings of the International Consensus on Periprosthetic Joint Infection. *Bone Joint J* 2013;95-B(11):1450-52. doi: 95-B/11/1450 [pii];10.1302/0301-620X.95B11.33135 [doi]
38. Weiss AP, Krackow KA. Persistent wound drainage after primary total knee arthroplasty. *J Arthroplasty* 1993;8(3):285-9. doi: 10.1016/s0883-5403(06)80091-4 [published Online First: 1993/06/01]
39. Southwood RT, Rice JL, McDonald PJ, et al. Infection in experimental hip arthroplasties. *J Bone Joint Surg Br* 1985;67(2):229-31. [published Online First: 1985/03/01]
40. Tande AJ, Osmon DR, Greenwood-Quaintance KE, et al. Clinical characteristics and outcomes of prosthetic joint infection caused by small colony variant staphylococci. *mBio* 2014;5(5):e01910-14. doi: 10.1128/mBio.01910-14 [published Online First: 2014/10/02]
41. Rakow A, Perka C, Trampuz A, et al. Origin and characteristics of haematogenous periprosthetic joint infection. *Clinical microbiology and infection : the official publication of the European Society of Clinical Microbiology and Infectious Diseases* 2019;25(7):845-50. doi: 10.1016/j.cmi.2018.10.010 [published Online First: 2019/01/27]
42. Trampuz A, Piper KE, Jacobson MJ, et al. Sonication of removed hip and knee prostheses for diagnosis of infection. *N Engl J Med* 2007;357(7):654-63. doi: 10.1056/NEJMoa061588 [published Online First: 2007/08/19]
43. Sayan A, Kopiec A, Shahi A, et al. The Expanding Role of Biomarkers in Diagnosing Infection in Total Joint Arthroplasty: A Review of Current Literature. *Arch Bone Jt Surg* 2021;9(1):33-43. doi: 10.22038/abjs.2020.42989.2169 [published Online First: 2021/03/30]
44. McNally M, Sousa R, Wouthuyzen-Bakker M, et al. The EBJIS definition of periprosthetic joint infection. *Bone Joint J* 2021;103-B(1):18-25. doi: 10.1302/0301-620X.103B1.BJJ-2020-1381.R1 [published Online First: 2021/01/01]
45. Zimmerli W, Moser C. Pathogenesis and treatment concepts of orthopaedic biofilm infections. *FEMS Immunol Med Microbiol* 2012;65(2):158-68. doi: 10.1111/j.1574-695X.2012.00938.x [doi]

46. Masters JP, Smith NA, Foguet P, et al. A systematic review of the evidence for single stage and two stage revision of infected knee replacement. *BMC Musculoskelet Disord* 2013;14:222. doi: 10.1186/1471-2474-14-222 [published Online First: 2013/07/31]
47. Osmon DR, Berbari EF, Berendt AR, et al. Diagnosis and management of prosthetic joint infection: clinical practice guidelines by the Infectious Diseases Society of America. *Clin Infect Dis* 2013;56(1):e1-e25. doi: cis803 [pii];10.1093/cid/cis803 [doi]
48. Bernard L, Arvieux C, Brunschweiler B, et al. Antibiotic Therapy for 6 or 12 Weeks for Prosthetic Joint Infection. *N Engl J Med* 2021;384(21):1991-2001. doi: 10.1056/NEJMoa2020198 [published Online First: 2021/05/28]
49. Chaussade H, Uckay I, Vuagnat A, et al. Antibiotic therapy duration for prosthetic joint infections treated by Debridement and Implant Retention (DAIR): Similar long-term remission for 6 weeks as compared to 12 weeks. *International journal of infectious diseases : IJID : official publication of the International Society for Infectious Diseases* 2017;63:37-42. doi: 10.1016/j.ijid.2017.08.002 [published Online First: 2017/08/15]
50. Lora-Tamayo J, Euba G, Cobo J, et al. Short- versus long-duration levofloxacin plus rifampicin for acute staphylococcal prosthetic joint infection managed with implant retention: a randomised clinical trial. *International journal of antimicrobial agents* 2016;48(3):310-6. doi: 10.1016/j.ijantimicag.2016.05.021 [published Online First: 2016/08/16]
51. Bernard L, Legout L, Zurcher-Pfund L, et al. Six weeks of antibiotic treatment is sufficient following surgery for septic arthroplasty. *The Journal of infection* 2010;61(2):125-32. doi: 10.1016/j.jinf.2010.05.005 [published Online First: 2010/06/15]
52. Zimmerli W, Widmer AF, Blatter M, et al. Role of rifampin for treatment of orthopedic implant-related staphylococcal infections: a randomized controlled trial. Foreign-Body Infection (FBI) Study Group. *JAMA* 1998;279(19):1537-41. doi: joc71805 [pii]
53. Aydin O, Ergen P, Ozturan B, et al. Rifampin-accompanied antibiotic regimens in the treatment of prosthetic joint infections: a frequentist and Bayesian meta-analysis of current evidence. *Eur J Clin Microbiol Infect Dis* 2021;40(4):665-71. doi: 10.1007/s10096-020-04083-4 [published Online First: 2020/10/31]
54. Nguyen S, Robineau O, Titecat M, et al. Influence of daily dosage and frequency of administration of rifampicin-levofloxacin therapy on tolerance and effectiveness in 154 patients treated for prosthetic joint infections. *Eur J Clin Microbiol Infect Dis* 2015;34(8):1675-82. doi: 10.1007/s10096-015-2404-z [doi]
55. Sensi P, Margalith P, Timbal MT. Rifomycin, a new antibiotic; preliminary report. *Farmaco Sci* 1959;14(2):146-7. [published Online First: 1959/02/01]
56. Bullock WE. Rifampin in the treatment of leprosy. *Rev Infect Dis* 1983;5 Suppl 3:S606-13. doi: 10.1093/clinids/5.supplement_3.s606 [published Online First: 1983/07/01]
57. Campbell EA, Korzheva N, Mustaev A, et al. Structural mechanism for rifampicin inhibition of bacterial rna polymerase. *Cell* 2001;104(6):901-12. doi: 10.1016/s0092-8674(01)00286-0 [published Online First: 2001/04/06]
58. Achermann Y, Eigenmann K, Ledergerber B, et al. Factors associated with rifampin resistance in staphylococcal periprosthetic joint infections (PJI): a matched case-control study. *Infection* 2013;41(2):431-37. doi: 10.1007/s15010-012-0325-7 [doi]

59. Acocella G, Brumfitt W, Hamilton-Miller JM. Evidence that rifampicin can be used safely for non-tuberculous diseases. *Thorax* 1980;35(10):788-91. doi: 10.1136/thx.35.10.788 [published Online First: 1980/10/01]
60. Lobo MC, Mandell GL. The effect of antibiotics on Escherichia coli ingested by macrophages. *Proc Soc Exp Biol Med* 1973;142(3):1048-50. doi: 10.3181/00379727-142-37173 [published Online First: 1973/03/01]
61. Reiter KC, Sambrano GE, Villa B, et al. Rifampicin fails to eradicate mature biofilm formed by methicillin-resistant Staphylococcus aureus. *Rev Soc Bras Med Trop* 2012;45(4):471-4. [published Online First: 2012/08/30]
62. Conlon BP, Nakayasu ES, Fleck LE, et al. Activated ClpP kills persisters and eradicates a chronic biofilm infection. *Nature* 2013;503(7476):365-70. doi: 10.1038/nature12790 [published Online First: 2013/11/15]
63. Tshefu K, Zimmerli W, Waldvogel FA. Short-term administration of rifampin in the prevention or eradication of infection due to foreign bodies. *Rev Infect Dis* 1983;5 Suppl 3:S474-S80.
64. Zimmerli W, Sendi P. Role of Rifampin against Staphylococcal Biofilm Infections In Vitro, in Animal Models, and in Orthopedic-Device-Related Infections. *Antimicrob Agents Chemother* 2019;63(2) doi: 10.1128/AAC.01746-18 [published Online First: 2018/11/21]
65. Lucet JC, Herrmann M, Rohner P, et al. Treatment of experimental foreign body infection caused by methicillin-resistant Staphylococcus aureus. *Antimicrob Agents Chemother* 1990;34(12):2312-7. doi: 10.1128/aac.34.12.2312 [published Online First: 1990/12/01]
66. Nessi R, Fowst G. Clinical use of rifampicin in combination for non-mycobacterial infections: a survey of published evidence. *J Int Med Res* 1979;7(3):179-86. doi: 10.1177/030006057900700304 [published Online First: 1979/01/01]
67. Norden CW. Experimental osteomyelitis. IV. Therapeutic trials with rifampin alone and in combination with gentamicin, sisomicin, and cephalothin. *J Infect Dis* 1975;132(5):493-9. doi: 10.1093/infdis/132.5.493 [published Online First: 1975/11/01]
68. Van der Auwera P, Klastersky J, Thys JP, et al. Double-blind, placebo-controlled study of oxacillin combined with rifampin in the treatment of staphylococcal infections. *Antimicrob Agents Chemother* 1985;28(4):467-72. doi: 10.1128/AAC.28.4.467 [published Online First: 1985/10/01]
69. Widmer AF, Gaechter A, Ochsner PE, et al. Antimicrobial treatment of orthopedic implant-related infections with rifampin combinations. *Clin Infect Dis* 1992;14(6):1251-53.
70. Wilson MG, Kelley K, Thornhill TS. Infection as a complication of total knee-replacement arthroplasty. Risk factors and treatment in sixty-seven cases. *J Bone Joint Surg Am* 1990;72(6):878-83. [published Online First: 1990/07/01]
71. Brandt CM, Sistrunk WW, Duffy MC, et al. Staphylococcus aureus prosthetic joint infection treated with debridement and prosthesis retention. *Clin Infect Dis* 1997;24(5):914-9. doi: 10.1093/clinids/24.5.914 [published Online First: 1997/05/01]
72. Karlsen OE, Borgen P, Bragnes B, et al. Rifampin combination therapy in staphylococcal prosthetic joint infections: a randomized controlled trial. *J Orthop Surg Res* 2020;15(1):365. doi: 10.1186/s13018-020-01877-2 [published Online First: 2020/08/30]
73. Morata L, Senneville E, Bernard L, et al. A Retrospective Review of the Clinical Experience of Linezolid with or Without Rifampicin in Prosthetic Joint Infections Treated with Debridement and Implant Retention. *Infect Dis Ther* 2014;3(2):235-43. doi: 10.1007/s40121-014-0032-z [published Online First: 2014/08/21]

74. Wouthuyzen-Bakker M, Tornero E, Morata L, et al. Moxifloxacin plus rifampin as an alternative for levofloxacin plus rifampin in the treatment of a prosthetic joint infection with Staphylococcus aureus. *International journal of antimicrobial agents* 2018;51(1):38-42. doi: 10.1016/j.ijantimicag.2017.04.011 [published Online First: 2017/07/03]
75. Parvizi J, Azzam K, Ghanem E, et al. Periprosthetic infection due to resistant staphylococci: serious problems on the horizon. *Clin Orthop Relat Res* 2009;467(7):1732-9. doi: 10.1007/s11999-009-0857-z [published Online First: 2009/05/02]
76. Lizaur-Utrilla A, Gonzalez-Parreno S, Gil-Guillen V, et al. Debridement with prosthesis retention and antibiotherapy vs. two-stage revision for periprosthetic knee infection within 3 months after arthroplasty: a case-control study. *Clinical microbiology and infection : the official publication of the European Society of Clinical Microbiology and Infectious Diseases* 2015;21(9):851 e11-7. doi: 10.1016/j.cmi.2015.05.028 [published Online First: 2015/06/07]
77. Bouaziz A, Uckay I, Lustig S, et al. Non-compliance with IDSA guidelines for patients presenting with methicillin-susceptible Staphylococcus aureus prosthetic joint infection is a risk factor for treatment failure. *Med Mal Infect* 2018;48(3):207-11. doi: 10.1016/j.medmal.2017.09.016 [published Online First: 2017/11/11]
78. Hirsiger S, Betz M, Stafylakis D, et al. The Benefice of Mobile Parts' Exchange in the Management of Infected Total Joint Arthroplasties with Prosthesis Retention (DAIR Procedure). *J Clin Med* 2019;8(2) doi: 10.3390/jcm8020226 [published Online First: 2019/02/13]
79. Geurts JA, Janssen DM, Kessels AG, et al. Good results in postoperative and hematogenous deep infections of 89 stable total hip and knee replacements with retention of prosthesis and local antibiotics. *Acta Orthop* 2013;84(6):509-16. doi: 10.3109/17453674.2013.858288 [published Online First: 2013/11/01]
80. Holmberg A, Thorhallsdottir VG, Robertsson O, et al. 75% success rate after open debridement, exchange of tibial insert, and antibiotics in knee prosthetic joint infections. *Acta Orthop* 2015:1-6. doi: 10.3109/17453674.2015.1026756 [doi]
81. Vilchez F, Martinez-Pastor JC, Garcia-Ramiro S, et al. Outcome and predictors of treatment failure in early post-surgical prosthetic joint infections due to Staphylococcus aureus treated with debridement. *Clin Microbiol Infect* 2011;17(3):439-44. doi: S1198-743X(14)63881-7 [pii];10.1111/j.1469-0691.2010.03244.x [doi]
82. Scheper H, Gerritsen LM, Pijls BG, et al. Outcome of Debridement, Antibiotics, and Implant Retention for Staphylococcal Hip and Knee Prosthetic Joint Infections, Focused on Rifampicin Use: A Systematic Review and Meta-Analysis. *Open Forum Infect Dis* 2021;8(7):ofab298. doi: 10.1093/ofid/ofab298 [published Online First: 2021/07/15]
83. Wouthuyzen-Bakker M, Sebillotte M, Lomas J, et al. Clinical outcome and risk factors for failure in late acute prosthetic joint infections treated with debridement and implant retention. *The Journal of infection* 2019;78(1):40-47. doi: 10.1016/j.jinf.2018.07.014 [published Online First: 2018/08/10]
84. Tornero E, Morata L, Martinez-Pastor JC, et al. KLIC-score for predicting early failure in prosthetic joint infections treated with debridement, implant retention and antibiotics. *Clinical microbiology and infection : the official publication of the European Society of Clinical Microbiology and Infectious Diseases* 2015;21(8):786 e9-86 e17. doi: 10.1016/j.cmi.2015.04.012 [published Online First: 2015/05/12]

85. Lora-Tamayo J, Euba G, Ribera A, et al. Infected hip hemiarthroplasties and total hip arthroplasties: Differential findings and prognosis. *The Journal of infection* 2013;67(6):536-44. doi: 10.1016/j.jinf.2013.07.030 [published Online First: 2013/08/13]
86. Buller LT, Sabry FY, Easton RW, et al. The preoperative prediction of success following irrigation and debridement with polyethylene exchange for hip and knee prosthetic joint infections. *J Arthroplasty* 2012;27(6):857-64 e1-4. doi: 10.1016/j.arth.2012.01.003 [published Online First: 2012/03/10]
87. Lowik CAM, Parvizi J, Jutte PC, et al. Debridement, antibiotics and implant retention is a viable treatment option for early periprosthetic joint infection presenting more than four weeks after index arthroplasty. *Clin Infect Dis* 2019 doi: 10.1093/cid/ciz867 [published Online First: 2019/09/11]
88. San Juan R, Garcia-Reyne A, Caba P, et al. Safety and efficacy of moxifloxacin monotherapy for treatment of orthopedic implant-related staphylococcal infections. *Antimicrob Agents Chemother* 2010;54(12):5161-6. doi: 10.1128/AAC.00027-10 [published Online First: 2010/09/22]
89. Scheper H, van Hooven D, van de Sande M, et al. Outcome of acute staphylococcal prosthetic joint infection treated with debridement, implant retention and antimicrobial treatment with short duration of rifampicin. *The Journal of infection* 2018;76(5):498-500. doi: 10.1016/j.jinf.2018.01.009 [published Online First: 2018/02/08]
90. Lesens O, Ferry T, Forestier E, et al. Should we expand the indications for the DAIR (debridement, antibiotic therapy, and implant retention) procedure for Staphylococcus aureus prosthetic joint infections? A multicenter retrospective study. *Eur J Clin Microbiol Infect Dis* 2018 doi: 10.1007/s10096-018-3330-7 [published Online First: 2018/08/08]
91. Pijls BG, Sanders I, Kuijper EJ, et al. Segmental induction heating of orthopaedic metal implants. *Bone Joint Res* 2018;7(11):609-19. doi: 10.1302/2046-3758.711.BJR-2018-0080.R1 [published Online First: 2018/12/26]
92. de Breij A, Riool M, Cordfunke RA, et al. The antimicrobial peptide SAAP-148 combats drug-resistant bacteria and biofilms. *Science translational medicine* 2018;10(423) doi: 10.1126/scitranslmed.aan4044 [published Online First: 2018/01/13]



Part I. The use of E-health to detect prosthetic joint infections

CHAPTER 2

A mobile app for postoperative wound care after arthroplasty: ease of use and perceived usefulness

Henk Scheper, Roxanne Derogee. Rachid Mahdad, Robert J.P. van der Wal, Rob G.H.H. Nelissen, Leo G. Visser, Mark G.J. de Boer

International Journal of Medical Informatics (2019); 129:75–80

Abstract

Background

Early postoperative discharge after joint arthroplasty may lead to decreased wound monitoring. A mobile woundcare app with an integrated algorithm to detect complications may lead to improved monitoring and earlier treatment of complications. In this study, the ease of use and perceived usefulness of such a mobile app was investigated.

Objective

Primary objective was to investigate the ease of use and perceived usefulness of using a woundcare app. Secondary objectives were the number of alerts created, the amount of days the app was actually used and patient-reported wound infection.

Methods

Patients that received a joint arthroplasty were enrolled in a prospective cohort study. During 30 postoperative days, patients scored their surgical wound by daily answering of questions in the app. An inbuilt algorithm advised patients to contact their treating physician if needed. On day 15 and day 30, additional questionnaires in the app investigated ease of use and perceived usefulness.

Results

Sixty-nine patients were included. Median age was 68 years. Forty-one patients (59.4%) used the app until day 30. Mean grade for ease of use (on a Likert-scale of 1 to 5) were 4.2 on day 15 and 4.2 on day 30; grades for perceived usefulness were 4.1 on day 15 and 4.0 on day 30. Out of 1317 days of app use, an alert was sent to patients on 29 days (2.2%). Concordance between patient-reported outcome and physician-reported outcome was 80%.

Conclusions

Introduction of a woundcare app with an alert communication on possible wound problems resulted in a high perceived usefulness and ease of use. Future studies will focus on validation of the algorithm and the association between postoperative wound leakage and the incidence of prosthetic joint infection.

Introduction

A prosthetic joint infection (PJI) is a feared complication for patients with a total joint arthroplasty. The reported incidence of PJIs ranges between 0.5-1.0% and 0.5-2.0% for hip and knee arthroplasty, respectively. This incidence is largely underestimated due to inadequate registration of infections[1]. Inadequate treatment of wound complications results in hospital readmission, revision surgery, long term antibiotic treatment and, in the worst case, removal of the prosthesis[2]. In The Netherlands, most patients are discharged the first or second postoperative day after arthroplasty, which is associated with faster functional recovery and lower costs[3]. Consequently, patients are responsible for monitoring their post-operative wound at home. This put them at risk for a delayed diagnosis of wound infections. This delay may lead to chronic PJI with extensive revision surgery with removal of the implant[4].

A mobile woundcare app used by patients after joint arthroplasty underscores the importance of adequate wound monitoring. Daily revision of the wound by patients may lead to improved monitoring, increased awareness for complications and, consequently, earlier consultation of the treating physician. There is evidence for distant postdischarge monitoring of postoperative patients. Reports have shown that post-operative telephone review is cost-effective and acceptable for patients with no underreporting of complications[5,6]. Another report showed a significant reduction in unnecessary emergency room visits by using email with smartphone photography in post-hypospadia patients[7]. The use of smartphones for monitoring recovery in post-operative patients at home has been shown to be feasible and acceptable to patients and surgeons, although patients were concerned about the lack of timely responses from healthcare[8,10]. To the best of our knowledge, no studies have been performed yet in which a mobile woundcare app was used with an integrated alert system for patients when to contact their physician. We hypothesized that a mobile woundcare app after joint implantation is useful for patients. We hypothesized that using such an app may lead to increased patient involvement, early detection of wound problems and prevention of chronic PJI[9].In this prospective study we investigated the ease of use and perceived usefulness of using such a mobile woundcare app in patients after joint arthroplasty.

Methods

All patients having a primary or revision total joint arthroplasty during the period July to December 2017 were eligible for participation in a prospective cohort study conducted at an academic hospital (Leiden University Medical Center) and a large regional teaching hospital (Alrijne Hospital). The primary objective was to investigate the ease of use and the patient's perceived usefulness of the woundcare app. Secondary objectives were the number of alerts, the number of calls to the treating physician during the study period, the amount of days the app was actually used, patient-reported wound infection and the concordance between patient-reported outcome and physician-reported outcome. The study was approved by the institutional Medical Ethical Committee (protocol nr. P17.091).

All patients scheduled for total joint arthroplasty were asked to participate during their hospital admission. Inclusion criteria were at least 18 years old, able to provide written informed consent and ownership of an android or iOS 9.0-or newer smartphone. Informed consent was obtained by the study coordinator who also guided each patient with downloading of the app. Instructions were given to patients how to use the app and how to fill in the daily review tasks. The study coordinator was available for the first 2-3 postoperative days for practical assistance and could be called during the study if needed. People who were unable to understand or read Dutch were excluded. After 30 days, patient files were reviewed to check for concordance between patient-reported and physician-reported outcome with respect to wound complications. All patients were seen in the outpatient clinic two and six weeks postoperatively. Clinicians were instructed about the underlying algorithm in the app and the alert system that could prompt patients to call them. It was left to the judgment of the treating clinicians to decide whether patients needed a clinical review or that a telephonic review was sufficient. The nurses on the ward were instructed about postoperative use of the app so they could help patients with filling in. Statistical analysis was done using SPSS (IBM SPSS Statistics version 24.0, Armonk, USA).

Mobile woundcare app

A woundcare app (figure 1) was developed by a digital innovation company (Innovattic, Delft, The Netherlands) with intellectual input from the authors.

All data entered in the app were pseudonomised and stored on a local ISO 27001 certified data management server at the coordinating hospital. A key for disclosure was stored on a local data safety folder. The app consisted of an introductory page collecting basic patient characteristics followed by daily short questionnaires regarding the patient's wound. Patients recorded redness, pain (by visual analogue score, VAS), wound leakage, fever and a picture of the wound could be taken (Appendix 1). After 30 days, the patient-

reported outcome was scored by the patient (i.e. PJI). Based on the daily questionnaires, an algorithm created daily a risk-score. A threshold score, developed by consensus meetings of the authors (HS, MB, RG, LV) defined above which the wound was thought to be at risk for being infected (Appendix 2). If the score exceeded this threshold, an alert message on the smartphone advised patients to contact their treating physician within 24 hours. The orthopaedic ward could be called directly via a push button in the app. Prior to the study, caregivers were instructed to register every contact in the electronic patient files. Apart from using the app, postoperative wound care did not differ between study participants and patients who were not included.

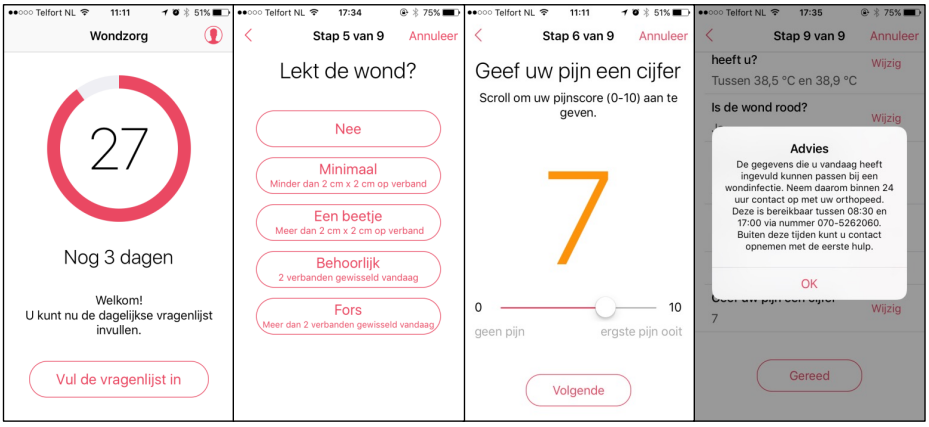


Figure 1. Screenshots of the woundcare app (with Dutch language)

English translation. Screen 1: Woundcare. Three days to go. You can now fill in the daily questionnaire. Screen 2: Does the wound leak? No, minimal (less then 2x2cm on the bandage), a little (more than 2x2cm on the bandage), fair (exchange of two bandages), strong (exchange of more than two bandages). Screen 3: Give your pain a score (Visual Analogue score 0-10). Screen 4. Advice: Your scores of today may fit with a wound complication. We advise you to contact your orthopaedic surgeon within 24 hours or (if out-of-office hours) with the emergency department

Ease of use and perceived usefulness

The questionnaires that were used to test for perceived usefulness and ease of use (Likert scale) were adapted from questionnaires that were developed for user acceptance of information technology[10] (Appendix 3). The app provided a link to the online questionnaires on day 15 and day 30 of the study. Additionally, patients received a reminder for the questionnaire by email. Responses followed a 5-point Likert scale from "strongly agree" to "strongly disagree." Results of day 15 and day 30 were compared for both questionnaires with a paired-samples T test. Patients who did not manage to fill in one of the questionnaires were contacted by telephone after 30 days to grade the app and to explore the reasons for not filling in the questionnaire.

Results

Of 127 eligible patients, thirty patients (24%) did not own a smartphone. Of the remaining 97 patients, 69 patients (71%) were included (Figure 1).

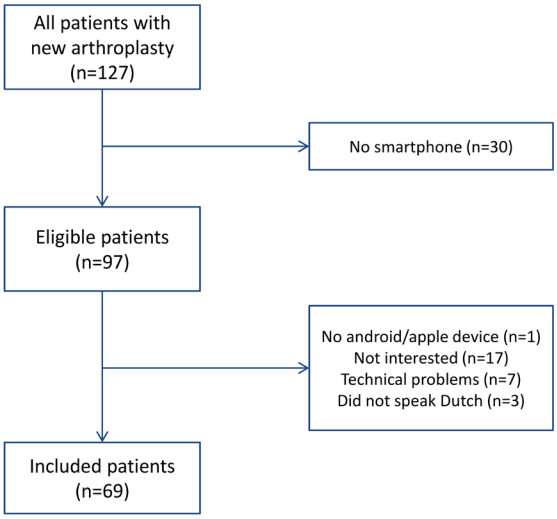


Figure 2. Selection and inclusion of patients.

Baseline characteristics of the included patients are shown in Table 1.

Table 1. Baseline characteristics of 69 patients who used the Woundcare app

Age (median, range)	68 (33-90)
Female/Male	46/23
University Medical Center (n)	19 (28%)
Regional hospital (n)	50 (72%)
Operating System Mobile Device	
iOS (n, %)	33 (48%)
Android (n, %)	36 (52%)
Joint arthroplasty	
Hip	32 (46%)
Knee	37 (54%)
Past medical history	
Diabetes mellitus	9 (13%)
Rheumatoid arthritis	8 (12%)
Megaprosthesis	2 (3%)

The median age was 68 years (range 33-90). Forty-one patients (59.4%) used the app until day 30. Nine patients (13.0%) stopped using the app immediately after the first or the second day of use. On average, the app was used by 43 patients per day. In total, the app was used on 1317 postoperative days (64% of the total amount of 30 postoperative days in 69 patients). The overall amount of responses tended to decline slowly over time (Figure 3).

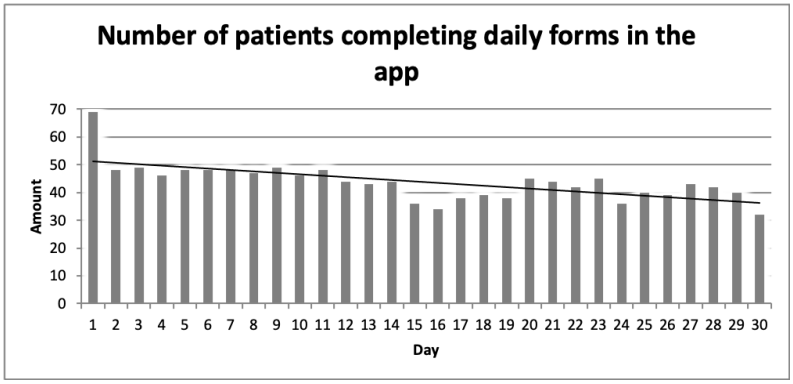


Figure 3. Number of patients completing daily forms in the app.

Perceived ease of use and perceived usefulness

The additional questionnaires about ease of use and usefulness were filled in by 31 patients (44.9%) on day 15 and by 37 patients (53.6%) on day 30. Fifteen patients (21.7%) filled in both questionnaires. The mean score for ease of use at day 15 was 4.2 (on a scale of 1 to 5) and 4.1 for perceived usefulness (Figure 4 and 5).

Figure 4. Patient-reported ease of use on day 15 and day 30

Mean scores (range)^	Day 15 (n=31)	Day 30 (n=37)
	Mean (range)	Mean (range)
Daily entry is easy	4.7 (3-5)	4.5 (4-5)
Questions are understandable	4.7 (4-5)	4.5 (4.5)
Size of text is right	4.6 (4-5)	4.4 (4.5)
App takes little time	4.3 (1-5)	4.4 (2-5)
Alerts are understandable*	4.3 (3-5)	3.9 (3.5)
Easy to fill in every day	4.2 (2-5)	4.1 (2-5)
Help the first day is useful	3.1 (1-5)	3.6 (1-5)
Questions easy to understand	4.5 (4-5)	4.5 (3.5)
App looks good	3.8 (2-5)	3.9 (3-5)
Daily reminder is useful	4.0 (1-5)	3.8 (1-5)
App is easy to use	4.4 (2-5)	4.3 (3-5)

* 16 patients scored 'not applicable'

^Scores: 1. strongly disagree, 2. disagree , 3. neutral, 4. agree , 5. strongly agree

Figure 5. Patient-reported perceived usefulness on day 15 and day 30

Mean scores (range)^	Day 15 (n=31)	Day 30 (n=37)
	Mean (range)	Mean (range)
Using app feels safe	4.2 (2-5)	4.1 (3-5)
Feels more engaged with app	4.0 (3-5)	3.9 (3-5)
Feels more responsible for recovery	3.7 (2-5)	3.8 (1-5)
Feels more engaged with hospital	3.8 (3-5)	3.7 (2-5)
Clear why app has been used	4.2 (3-5)	4.2 (4-5)
Feels more in control of wound	3.8 (2-5)	3.8 (2-5)
App is useful	4.3 (3-5)	4.2 (3-5)
App does not give stress	4.5 (3-5)	4.4 (3-5)
Sensible to fill in every day	4.1 (2-5)	4.0 (1-5)
Alerts are useful	4.1 (2-5)	4.2 (2-5)
Recommend app	4.4 (2-5)	4.2 (3-5)
Hospital took calls seriously*	3.7 (3-5)	3.6 (3-5)

* 16 patients scored 'not applicable'

^Scores: 1. strongly disagree, 2. disagree , 3. neutral, 4. agree , 5. strongly agree

The scores on day 30 were comparable to day 15 for ease of use (score 4.2, p=0.43) and perceived usefulness (score 4.0, p=0.40). The average satisfaction with the app at day 15 was 8.2 (on a scale of 1 to 10; range 6 to 10). Sixteen patients (23%) who did not fill in a questionnaire at all were contacted by telephone, to have information on user-friendliness or hick-ups when using the app. Eight of them could be reached and were interviewed with predefined questions. The mean satisfaction-score of the app among them was 7.9 (range 7-10). The majority of these patients had stopped using the app prior to reaching the day of the questionnaire (day 15). Reasons for discontinuation were malfunction of the smartphone (n=1), the app had stopped giving reminders (n=2) or patients had forgotten to fill in the app (n=6).

Alerts

An alert was sent to patients on 29 (2.2%) of the 1317 days the app was used. Ten alerts were sent because the score exceeded 5 points, three alerts because the score exceeded four points on two consecutive days and 16 alerts because the score exceeded three points on three consecutive days (see also Appendix 2). Thirteen patients responded on the question of the online questionnaire specifically asking if the hospital took their calls, based on alerts, seriously (score 3.7 on day 15 and 3.6 on day 30, figure 4). No single record of patient calls was found in the electronic patient files. Also, it appeared that in the iOS version of the app there was a technical flaw in the algorithm resulting in only sending alerts when the score exceeded five points. Due to this flaw, 28 out of 57 alerts were not sent to the patient.

Postoperative course

Forty-one patients filled in the outcome score on complications on day 30. Concordance of patient-reported and physician-reported outcome was reached in 33 patients (80%) (Table 2).

Table 2. Concordance between patient-reported and physician-reported outcome in 41 patients who used the app until day 30

		Physician-reported outcome			
		I don't know	No infection	Suspicion PJI	PJI
Patient-reported outcome	I don't know	0	7	0	0
	No infection	0	33	0	0
	Suspicion PJI	0	0	0	1
	PJI	0	0	0	0

Discordance occurred in seven patients who did not have a complication, but scored "I don't know" as outcome. The only patient (1.5%) in our study that developed a PJI on day 30 scored a "suspected PJI, but appeared to be no infection".

One patient (1.5%) had revision surgery because of repeated dislocations of the hip joint. Two patients (2.9%) developed a deep venous thrombosis of the leg. Four patients (5.8%) reported a temperature >38.0°C at least once during the 30 day postoperative period. Postoperative wound leakage was reported by thirty-seven patients (53.6%); the majority of the patients reported this on the second and third postoperative day (Figure 6).

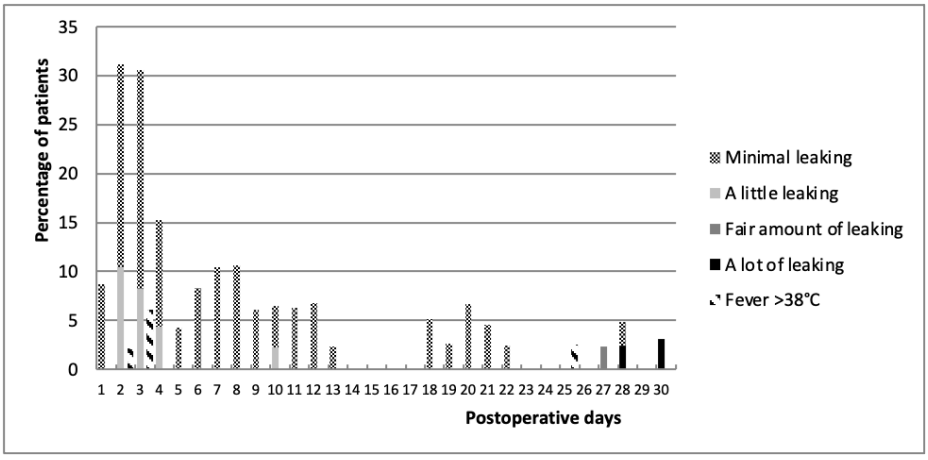


Figure 6. Proportion of patient-reported amount of wound leakage and fever (>38.0°C).

Wound leakage duration was present during a mean 2.2 days (range 1 to 11). From day 18 onwards, five patients reported new wound leakage for one to five days. The leakage reported in the fourth postoperative week corresponded with the patient who developed a prosthetic joint infection. This patient scored an unchanged wound for four weeks and leakage and fever since one day before admission with a PJI.

Discussion

We found that introduction of a mobile woundcare app resulted in a high perceived usefulness and ease of use. Patients felt engaged with their health and with the care provided by the hospital. This involvement was consistent during the use of the app. The number of patients completing daily forms in the app declined only mildly, confirming patient engagement with their own woundcare.

Clinical applications of mobile e-health by patients can be a valuable tool in health care management. With the increasing use of medical apps, it is important to develop e-tools that support patients and clinicians in improving health care. The high inclusion rate in this study stresses patient willingness to use mobile apps for postoperative wound monitoring. This is in line with recent surveys showing that using an app for surgical wound monitoring, including taking digital wound photographs, is supported by most patients[11,12]. Of all eligible patients aged 65 years or more, smartphone ownership in this study was 76%. Most likely, this will increase over the next years resulting in more patients who may benefit from medical apps.

In our woundcare app postoperative follow up care by patients themselves is integrated with an (wound)risk assessment that supports the patient when to contact their physician. Other studies have suggested that the use of mobile e-health led to more engagement of patients with their treatment[13-15]. Importantly, negative experiences might arise when daily asked to monitor a postoperative wound; however, these were not reported by patients. The response rate for the questionnaires on day 15 and 30 of only 77% might introduce a selection bias with skewed positive responses. Therefore, patients who did not fill in a questionnaire were interviewed later by telephone and, using the same grading system, those non-responders showed comparable high satisfaction rates as responders.

Cost-effectiveness

If postoperative infections can be treated at an earlier stage, devastating chronic PJI can be prevented. The costs of revision surgery for one patient (estimated costs around

30.000 euro) are about the same as the costs for the development of this app[16]. The app may be cost-effective by preventing diagnostic delay but larger studies need to be done to show cost-effectiveness. The app worked well for the only patient who developed a PJI; this patient scored eight points on the day of admission (score based on heavy leakage and a high pain score). She had not used the app on the day prior to admission; two days before admission her score was four. Good compliance is needed in order to really benefit from the app. Of all included patients, 59% used the app as intended until day 30. One of the main - understandable - reasons for discontinuation was that patients deemed further use of the App irrelevant, since their postoperative recovery went uneventful. For these patients, further use of the app would obviously not have resulted in improved clinical outcome.

Patient-reported and physician-reported outcome

Concordance between patient-reported and physician-reported outcome on wound healing is important in order to estimate the accuracy of patients to determine their own diagnosis. The discordance rate of 20% in this study is probably secondary to outcome options that were not presented clearly in the app. The 'I don't know' category (table 2) was too vague in hindsight and will be omitted in the next version of the app. We estimate that, when adjusting the options in the app, the concordance comes close to 100%, but reliable estimation of concordance can only be addressed in a larger study.

One of the objectives of this study was to determine the number of alerts that led to a call to the treating physician resulting in a change in treatment. The one patient that developed a PJI did receive an alert and was admitted to the hospital on the same day. Although physicians were instructed to report all app-based phone calls by patients in the electronic patient files, this apparently did not happen. This can partly be explained by the reduced number of alerts (due to the technical problems) but also by underreporting. The technical problems underscore the importance of pilot studies like this to find and resolve these issues. Visual integration of all app data into patient's electronic files may lead to improved registration, as this supports physicians to interpret a clinical situation more accurate when called by their patients. Currently, real-life visual integration of the clinical data of the app in the electronic patient files is implemented in our hospital.

Scoring system for wound infection

As far as we know, there is no validated grading system to score a postoperative wound. A systematic review of surgical infection scoring systems found one scoring system for postoperative sternal wounds, but this was developed for scoring by physicians and not suited for patient monitoring[17]. We developed a grading system based on the classical criteria for wound infection after arthroplasty (pain, fever, leakage, redness) that is easy

to use for patients (Appendix 2). To avoid false-negative results the threshold for sending an alert was put low, resulting in alerts in ten individual patients, while only one patient developed a PJI. Most of these alerts were based on a high VAS score; for these patients a mobile app may lower the threshold tot contact the treating physician to optimise their pain medication.

Wound leakage and infection

Currently, the importance of postoperative wound leakage as risk factor for PJI is largely unknown[16]. Differentiation between wound leakage as being part of normal postoperative course or being a symptom of a PJI is essential. Maathuis et al. reported that 10% of all wound leakages resulted in a PJI (unpublished results). Currently, a multicenter study on the treatment of postoperative wound leakage in elective hip and knee arthroplasty is done[16]. Immediate extensive surgical debridement is the cornerstone of treatment for an acute PJI but if done unnecessary it exposes patients to an additional risk for infection. Many patients in this study (59.4%) reported postoperative wound leakage, the majority on the second and third postoperative day. The true incidence of wound leakage may be higher, since not all patients completed the app every day. The recurrence of leakage on day 18 in five patients might be explained by easier wound monitoring after removal of the plaster at two weeks postoperative. This study was not powered for finding an association between the length and severity of wound leakage and a postoperative PJI. This association should be addressed in a large cohort study. A causal relationship would underscore the need for strict wound monitoring for which postoperative wound care with this app may have an additional value.

Funding

The app was funded by a grant by the department of Orthopaedics at the LUMC and by a grant of a healthcare insurance company Zorg & Zekerheid.

Acknowledgments

We are indebted to Barbara Snoeker who helped us with the preparation of the study in Alrijne Hospital

References

1. Gundtoft PH, Overgaard S, Schonheyder HC, Moller JK, Kjaersgaard-Andersen P, Pedersen AB. The "true" incidence of surgically treated deep prosthetic joint infection after 32,896 primary total hip arthroplasties: a prospective cohort study. Acta Orthop. 2015;86(3):326-34.
2. Widmer AF. New developments in diagnosis and treatment of infection in orthopedic implants. Clin Infect Dis. 2001;33 Suppl 2:S94-106.
3. Hansen TB. Fast track in hip arthroplasty. EFORT Open Rev. 2017;2(5):179-88.
4. Amlie E, Lerdal A, Gay CL, Hovik O, Nordsletten L, Dimmen S. A Trend for Increased Risk of Revision Surgery due to Deep Infection following Fast-Track Hip Arthroplasty. Adv Orthop. 2016;2016:7901953.
5. Gray RT, Sut MK, Badger SA, Harvey CF. Post-operative telephone review is cost-effective and acceptable to patients. Ulster Med J. 2010;79(2):76-9.
6. Armstrong KA, Coyte PC, Brown M, Beber B, Semple JL. Effect of Home Monitoring via Mobile App on the Number of In-Person Visits Following Ambulatory Surgery: A Randomized Clinical Trial. JAMA Surg. 2017;152(7):622-7.
7. Chua ME, Saunders MA, Bowlin PR, Ming JM, Lopes RI, Farhat WA, et al. Impact of smartphone digital photography, email, and media communication on emergency room visits post-hypospadias repair. Can Urol Assoc J. 2017;11(3-4):E134-E7.
8. Sanger PC, Hartzler A, Han SM, Armstrong CA, Stewart MR, Lordon RJ, et al. Patient perspectives on post-discharge surgical site infections: towards a patient-centered mobile health solution. PLoS One. 2014;9(12):e114016.
9. Kaur R, Lopez V, Thompson DR. Factors influencing Hong Kong Chinese patients' decision-making in seeking early treatment for acute myocardial infarction. Res Nurs Health. 2006;29(6):636-46.
10. Davis FD. Perceived Usefulness, Perceived Ease of Use, and User Acceptance of Information Technology. MIS Quarterly. 1989;13(3):319-40.
11. Wiseman JT, Fernandes-Taylor S, Barnes ML, Tomsejova A, Saunders RS, Kent KC. Conceptualizing smartphone use in outpatient wound assessment: patients' and caregivers' willingness to use technology. J Surg Res. 2015;198(1):245-51.
12. Abelson JS, Symer M, Peters A, Charlson M, Yeo H. Mobile health apps and recovery after surgery: What are patients willing to do? Am J Surg. 2017;214(4):616-22.
13. Fox BI, Felkey BG. Engaging patients with digital tools: what we think we know. Hosp Pharm. 2014;49(10):992-3.
14. Agboola S, Havasy R, Myint UK, Kvedar J, Jethwani K. The impact of using mobile-enabled devices on patient engagement in remote monitoring programs. J Diabetes Sci Technol. 2013;7(3):623-9.
15. Pavliscsak H, Little JR, Poropatich RK, McVeigh FL, Tong J, Tillman JS, et al. Assessment of patient engagement with a mobile application among service members in transition. J Am Med Inform Assoc. 2016;23(1):110-8.
16. Lowik CAM, Wagenaar FC, van der Weegen W, Poolman RW, Nelissen R, Bulstra SK, et al. LEAK study: design of a nationwide randomised controlled trial to find the best way to treat wound leakage after primary hip and knee arthroplasty. BMJ open. 2017;7(12):e018673.
17. A systematic review of surgical infection scoring systems used in surgical patients. JBI Libr Syst Rev. 2011;9(60):2627-83.

Appendix A.

Daily reviews to be filled in by patient in Woundcare App

Questions	Response options
Do you have a fever?	yes/no/not measured
How high is your fever?	<37.5 °C/37.5-37.9/38-38.4/38.5-38.9/ >39
Is the wound red?	yes/no/not judgeable
How is the redness compared to yesterday?	more red/less red/unchanged/not judgeable
Is the wound leaking?	no/ minimal (<2x2cm on dressing)/ a little (>2x2cm on dressing)/ fairly (2 dressing changes today)/ a lot (>2 dressing changes today)
Give your pain a number. (VAS score)	0-10

Questionnaires at day 30

Question	Response options
What was the date of discharge?	date
Have you had another surgery?	Yes/no
When did you have another surgery?	date
Did you receive antibiotics?	Yes/no
Which antibiotics did you receive?	
What was the diagnosis?	No infection / Superficial infection (Spontaneous recovery) / Superficial infection (Recovery with antibiotics) / PJI (operation and antibiotics received) / Suspected PJI (operation proved otherwise) / I don't know
Did you answer the previous question with your physician?	Yes/no

Appendix B.

Underlying algorithm in woundcare App in order to send alerts to patients with a possible postoperative complication

Question	Answer options	Points
Do you have a fever?	T > 38.5	5
	T 38-38.5	2
	T>2 days T 38-38.5	5
	T<38	0
Is the wound leaking?	No	0
	Minimal	1
	A little	2
	Fairly	3
	A lot	4
Is de wound red?	More red than day before	2
VAS score	VAS >7	4
	VAS 6 or 7	3
	VAS ≤ 5	0
	VAS > 2pts compared to day before	3
	When >3 days VAS>3	3
	Total amount of points:	

Woundcare App calculates amount of points per day:

- If score ≥ 5 points: an alert will appear on the smartphone: "the symptoms that you filled in on your app today might fit with a wound problem. We advise you to consult your orthopaedist within 24 hours."
- If for ≥ 2 consecutive days 4 points: same alert message on smartphone
- If for ≥3 consecutive days 3 points : same alert message on smartphone
- If none of above mentioned points: message: "thank you for using the app today, You can use the app tomorrow again."

Appendix C.

Questionnaires perceived ease of use and perceived usefulness

Perceived ease of use-questionnaire

1. Filling in the daily form of the App is easy for me
2. The questions are understandable
3. The sizing of the text in the App is right
4. Filling in the daily form takes a lot of time
5. If I received the advice to call my physician, it was clear to me what I had to do
6. I find it difficult to keep filling in the daily form
7. It is good to receive help with filling in the app on the first day
8. The questions in the App are difficult to understand
9. The design, in other words the look of the App is attractive
10. The daily reminder to fill in the App is useful
11. The use of the App is easy

Strongly disagree / disagree / neutral / agree / strongly agree

(answers question 5: Strongly disagree / disagree / neutral / agree / strongly agree / Does not apply)

Perceived usefulness -questionnaire

1. It feels safe to use this App to monitor my wound
2. I feel more involved in my woundcare by using the App
3. I feel more responsible for my own wound by using the App
4. I feel more involved with the hospital by using the App
5. It is clear to me why I need to use the App
6. Using the App makes me feel more in control of the wound
7. I think the Woundcare App is useful
8. Using the App makes me feel stressed
9. I think it is useless to fill in the daily forms every day
10. I think it's useful to get the advice to call the hospital at certain scores
11. I would recommend the App to others
12. I was taken seriously when the App advised me to call the hospital

Strongly disagree / disagree / neutral / agree / strongly agree

(answers question 12: Strongly disagree / disagree / neutral / agree / strongly agree / Does not apply)

What grade do you give the App?
(1 = very bad, 10 = very good)
1 2 3 4 5 6 7 8 9 10

Open question: What could be improved in the App according to you?

CHAPTER 3

Wound drainage after arthroplasty and prediction of acute prosthetic joint infection: prospective data from a multicenter cohort study using a telemonitoring app

Henk Scheper, Rachid Mahdad, Brenda Elzer, Claudia Löwik, Wierd Zijlstra, Taco Gosens, Joris C.T. van der Lugt, Robert J.P. van der Wal, Rudolf W. Poolman, Matthijs P. Somford, Paul C. Jutte, Pieter K. Bos, Richard E. Zwaan, Rob G.H.H. Nelissen, Leo G. Visser, Mark G.J. de Boer on behalf of the *wound care app study group**

Journal of Bone and Joint Infection 2023, 8, 59–70

Abstract

Background

Differentiation between uncomplicated and complicated postoperative wound drainage after arthroplasty is crucial to prevent unnecessary resurgery. Prospective data about the duration and amount of postoperative wound drainage in patients with and without PJI are currently absent.

Methods

A multicenter cohort study was conducted to assess the duration and amount of wound drainage in patients after arthroplasty. During 30 postoperative days after arthroplasty, patients recorded their wound status in a previously developed wound care app and graded the amount of wound drainage on a 5-point scale. Data about prosthetic joint infection (PJI) in the follow-up period were extracted from the patient files.

Results

Of 1019 included patients, 16 patients (1.6%) developed a PJI. Minor wound drainage decreased from the first to the fourth postoperative week from 50% to 3%. Both moderate to severe wound drainage in the third week and newly developed wound drainage in the second week after a week without drainage were strongly associated with PJI. (OR 103.23, 95% CI 26.08 to 408.57, OR 80.71, 95% CI 9.12 to 714.52, respectively). The positive predictive value for PJI was 83% for moderate to heavy wound drainage in the third week.

Conclusion

Moderate to heavy wound drainage and persistent wound drainage were strongly associated with PJI. The positive predictive value of wound drainage for PJI was high for moderate to heavy drainage in the third week but was low for drainage in the first week. Therefore, additional parameters are needed to guide the decision to reoperate patients for suspected acute PJI.

Introduction

Total hip and knee arthroplasties are highly successful treatment modalities for advanced osteoarthritis, the most common joint disorder worldwide [1]. A prosthetic joint infection (PJI), which develops in approximately 1-2% of all arthroplasties, is a serious and devastating postoperative complication with a high impact on a patient's well-being[2,3]. Postoperative wound drainage is frequently reported as an important indicator for the presence of PJI [4-6]. Wound drainage may be an early symptom of a present PJI but may also be a risk factor for subsequent development of PJI [5,7]. Discrimination between infectious and non-infectious postoperative wound drainage is of crucial importance. When the prosthetic joint is infected, surgical debridement and protracted antimicrobial treatment is required. For 'noninfectious' serosanguinous drainage caused by intraoperative disruption of soft tissue and capillaries, only conservative wound management is indicated [4].

In 2013, the first International Consensus Meeting (ICM) on PJI advised that surgical management of persistent wound drainage should be performed without delay if wound drainage persists for five to seven days after index surgery [8]. According to the recently published EBJIS definition for PJI, an history of prolonged wound drainage (as a feature of wound healing problem) is a clinical sign included in the PJI likely category [9]. However, these recommendations were not backed up by research data about duration of postoperative wound drainage as summarized in a recent systematic review [4]. Collecting wound drainage data is challenging because most patients are discharged from hospital soon after surgery. The use of smartphone applications for distant telemonitoring of postoperative patients has been shown to be feasible and acceptable for both patients and surgeons [10-13]. In an earlier study, the use of a postoperative wound care app that was developed at Leiden University Medical Center, showed a high perceived usefulness and ease of use as reported by patients[14]. To assess the amount and duration of postoperative wound drainage after joint arthroplasty in patients with and without PJI, we conducted a nationwide cohort study using this smartphone application in which we collected detailed information regarding the condition and natural history of the postoperative wound.

Methods

A multicenter, prospective observational study was conducted in 11 Dutch academic and non-academic hospitals between November 1st, 2019 and October 1st, 2021. All patients aged 18 years and older who received a knee or hip arthroplasty, who were able to provide informed consent, owned an android or iOS smartphone and were able to read Dutch language, were eligible for inclusion. Patients were screened during or after preoperative visits by a local

nurse specialist or the coordinating study nurse. Informed consent was obtained via the woundcare app. Instructions how to use the app were provided to all patients by the local research coordinator. The nurses on the ward as well as the study coordinator were available for help with the use of the app during admission and throughout the study. All patients received routine postoperative medical care in the outpatient clinic as per local protocol in each participating hospital. Primary endpoint was the extent and duration of postoperative wound drainage in patients with and without PJI. Secondary endpoints were the association between presence of self-reported fever, redness and pain and PJI, and the validation of the designed algorithm for sending alert messages for suspected PJI. PJI was defined according to the criteria from the European Bone and Joint Infection Society.[9]

The use and function of the app has been described previously [14]. In short, for 30 days following joint arthroplasty patients recorded their wound status daily on their mobile app. Redness, pain (by visual analogue score, VAS), wound drainage and presence of fever were recorded, and a picture of the wound could be taken. Based on the questionnaires, an inbuilt algorithm created a daily risk score (see Appendix A). If this score exceeded a predefined threshold, which was based on expert consensus of participating clinicians, an alert message was issued that allowed patients to contact their treating physician via a push button in the app. It was for the attending clinicians to decide whether patients needed a clinical review or not. If wound drainage during the first 14 days was not reported, patients were allowed to stop using the app. They were instructed to resume using the app if new drainage or other complications arose. After both 30 and 90 days, all patients were asked to report postoperative complications in the app. After a minimum follow up period of 90 postoperative days, endpoint data were extracted both from the app as from the electronic patient files to enable comparison of patient-reported and physician-reported outcome. If discordant, the outcome reported by the attending orthopedic surgeon was regarded as the final outcome.

The study was conducted according to the principles of the Declaration of Helsinki. The study was approved by the ethics review committees and a waiver was obtained to use electronic instead of written informed consent. The use of the app for this study was approved by the Dutch Health Inspectorate (reference number VGR2O1 1434). The app was developed by software company Innovattic. This company was not involved in the setup, data-analysis and report of this study.

Quantification of wound drainage

The International Consensus Meeting (ICM) on PJI defined persistent wound drainage as >2 × 2 cm of drainage in the wound dressing beyond 72 hours after index surgery. However, this definition lacks a more detailed quantification of wound drainage [15]. Therefore, we used a proposed classification of persistent wound drainage which is currently used in another

Dutch wound drainage study (National Trial Registration 5960)[16]. On a daily basis, the patient had to enter the following drainage scores in the app: no drainage, minimal drainage, mild drainage, moderate drainage or heavy drainage (for exact definitions, see Table 1).

Table 1. Self-reported wound characteristics by patients in the wound care app.

Characteristic	Daily available scores for the patient after surgery
Fever	T < 38°C
	T 38-38.5°C
	T > 38.5°C
Wound drainage	No
	Minimal: <2x2cm on bandage
	Mild: >2x2cm on bandage
	Moderate: 1-2 bandages exchanged
	Heavy: >2 bandages exchanged
	Not judgeable (e.g., due to plaster/wound dressing)
Redness of wound	No
	Yes, less red than yesterday
	Yes, same as yesterday
	Yes, increased compared to yesterday
	Not judgeable (e.g., due to plaster/wound dressing)
Pain score (Visual Analogue Score)	Score 0-10 (via a slider in the app)

Statistical analysis

Descriptive statistics were used for baseline characteristics. To address missing values of wound drainage, the most recent drainage score was carried forward if data were missing after the first 14 days but only if the most recent drainage score was 'no drainage'. The cut-off of 14 days was based on the recommendation of the app to stop using the app after 14 days and only reuse it if any new complications arose. Odds ratios, sensitivity, specificity, and negative and positive predictive values were calculated to examine the strength of the association between mild or moderate to heavy wound drainage and PJI and between duration of wound drainage and PJI. Median duration of wound drainage was compared between patients with and without PJI using Mann Whitney U Test. All statistical analyses were performed with SPSS (IBM SPSS Statistics version 25.0, Armonk, USA).

Data flow and management

Privacy-sensitive data entered into the app by patients were pseudonymized with trusted real-time encryption. Encryption keys and a list of investigators who were allowed for de-encryption were stored by a Trusted Third Party (ZorgTTP). The encryption code and the data entered in the app were sent to a research database and were only decrypted to review the physician-reported outcome. Data files used for analysis will be stored on a local safe network storage facility.

Results

Of all patients eligible for inclusion during the study period, 1019 patients were included (total hip arthroplasty 46%, total knee arthroplasty 54%). Baseline and outcome characteristics are summarized in Table 2. During the first two postoperative weeks, the app was used by more than 80% of patients per day (Figure 1). The app use declined during the third and fourth week from 80% to 30%, consistent with the advice that use of the app beyond two weeks was only needed if new drainage or other complications would occur.

Table 2. Baseline and outcome characteristics of 1019 patients as entered in the app#

	Reported by patient in app	Definite report by study team
Baseline characteristics		
Age (median, range)	65 (18-90)	n/a
BMI (mean, SD)	29.1 (11.0)	n/a
Type of joint arthroplasty		
Knee	467 (46%)	n/a
Hip	547 (54%)	n/a
Other (shoulder, ankle)	2 (0.2%)	n/a
Tumour prosthesis (n, %)	10 (1%)	n/a
Past medical history		
Diabetes mellitus	73 (7%)	n/a
Rheumatoid arthritis	60 (6%)	n/a
Report of outcome		
Prosthetic joint infection	16 (1.5%)	16 (1.6%)
Surgery for suspected PJI, appeared to be no PJI	5 (0.5%)	3 (0.3)
Superficial wound infection, resolved after antibiotic treatment	5 (0.5%)	6 (0.6%)
Superficial wound infection, spontaneously resolved	22 (2.2%)	2 (0.2%)
No data available	176 (17.5%)*	39 (3.8%)^
I don't know	121 (11.9)	-
No complication (if data available)	674/843 (80.0%)	956/980 (97.6%)

#Outcome checked until three months postoperative

*179 patients did not fill out the outcome after 30 and 90 days.

^From one study center, data from the 39 included patients could not be retrieved from the local researcher.

n/a: not applicable

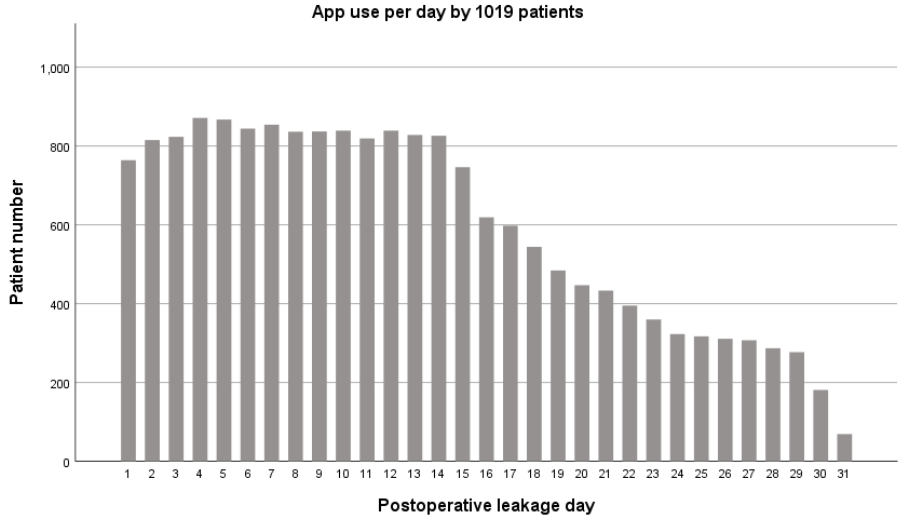


Figure 1. Daily wound care app use by patients during postoperative period

The incidence of postoperative wound drainage in patients with and without PJI is reported in Table 3 and Figure 2. During the first, second, third and fourth postoperative week, any form of wound drainage was present in 50%, 12%, 8% and 3% of patients without PJI and in 63%, 88%, 64% and 25% of patients with PJI. The high proportion of drainage in the first week was predominantly caused by minimal leakage (defined as <2x2cm on gauze) occurring in 87% (424/489) of patients without PJI. In this group, 51 patients (5%) had moderate to heavy wound drainage in the first week, decreasing to 1%, 1% and 0.1% in the next weeks. Moderate to heavy wound drainage of patients with PJI occurred in 25%, 38%, 46% and 0% of patients during four weeks. Reported redness (10%), fever (5%) and high pain scores (VAS >7, 11%) were mainly reported during the first week and declined thereafter. Proportions of wound drainage in patients without PJI varied depending on the type of joint, BMI and the presence of diabetes (Table 4).

Sixteen (1.6%) patients developed a PJI during the follow up period. Fourteen patients experienced an early postoperative PJI after a median of 14 days (IQR 10-18days). Two patients developed an early chronic PJI on postoperative day 71 and 77 (Table 5). Three patients were reoperated for a suspected PJI that was subsequently not confirmed (e.g. hematoma). Six patients (0.6%) received a short course of antibiotics for a presumed superficial wound infection but did not develop a PJI. The strongest risk factors for PJI were any wound drainage in the second week (OR 50.83, 95% CI 11.41-226.51), moderate to heavy drainage in the second (OR 51.22, 95% CI 15.84-165.65) or third week (OR 103.23, 95% CI 26.08-408.57). New onset drainage in the second week after a week without

Table 3. Reported postoperative wound drainage in all patients with and without prosthetic joint infection.

	1st week	
	No PJI	**PJI**
App use per week (n patients)	978	16
Wound drainage		
No wound drainage at all during week	416 (43%)	5 (31%)
Any wound drainage anywhere during week	489 (50%)	10 (63%)
Minimal (<2x2cm on gauze)	424 (87%)	8 (80%)
Mild (>2x2cm on gauze)	181 (37%)	7 (70%)
Moderate (1-2 gauzes exchanged)	41 (8%)	4 (40%)
Heavy (>2 gauzes exchanged)	10 (2%)	-
New onset drainage after 1week no drainage	-	-
> 4 days of wound drainage during week	82 (8%)	3 (19%)
Drainage not assessable*	165 (17%)	1 (6%)
Redness		
Any wound redness during week	100 (10%)	3 (19%)
Increased redness	32 (32%)	1 (33%)
Fever		
Fever during postoperative period	53 (5%)	-
Pain		
VAS > 5 anytime during week	360 (37%)	5 (33%)
VAS > 7 anytime during week	107 (11%)	0 (0%)
Alerts		
Any alerts during week	415 (42%)	8 (53%)
Number of alerts per week per patient		

*Patients with or without any drainage who could not assess wound drainage during one or more days during week due to gauzes in situ.

drainage (OR 80.71, 95% CI 9.12-714.52) and more than 5 cumulative wound drainage days during the first three postoperative weeks (OR 9.20, 95% CI 3.37-25.14) were also strongly associated with development of PJI (Table 6). Drainage for more than five days during the first three weeks predicted PJI with sensitivity of 63% and specificity of 87%, while drainage for more than 10 days predicted PJI with sensitivity of 27% and specificity of 97% (Appendix B). No wound drainage at all was reported by 467 patients (46%). Of them, only one patient developed a PJI resulting in a negative predictive value of no wound drainage as indicator for recovery without PJI of >98% (Table 6). The positive predictive value of any amount of wound drainage for PJI was low during the four postoperative weeks (2%, 11%, 8% and 4%, respectively) and increased for moderate-heavy wound drainage, especially in the third postoperative week (8%, 35%, 83%, 0%, respectively). Over the 4-week postoperative period, the average number of alerts per patient was not higher for patients with PJI compared to

2nd week		3rd week		4th week	
No PJI	PJI	No PJI	PJI	No PJI	PJI
950	16	999	11	999	4
789 (83%)	2 (13%)	903 (90%)	4 (36%)	973 (97%)	3 (75%)
115 (12%)	14 (88%)	76 (8%)	7 (64%)	25 (3%)	1 (25%)
98 (85%)	12 (86%)	65 (86%)	3 (43%)	24 (96%)	1 (100%)
25 (22%)	5 (36%)	19 (25%)	3 (43%)	4 (16%)	1 (100%)
10 (9%)	6 (43%)	1 (1%)	5 (71%)	1 (4H%)	-
2 (2%)	2 (14%)	-	-	-	-
28 (5%)	2 (13%)	25 (5%)	2 (50%)	4 (1%)	1 (25%)
31 (3%)	4 (25%)	11 (1.1%)	1 (9%)	4 (0,4%)	1 (25%)
50 (5%)	-	23 (2%)	-	1 (0.1%)	-
45 (5%)	3 (19%)	37 (4%)	3 (19%)	20 (2%)	0 (0%)
20 (44%)	2 (66%)	9 (24%)	0 (0%)	6 (30%)	0 (0%)
21 (2%)	1 (6%)	12 (1%)	2 (18%)	5 (0,5%)	1 (25%)
114 (12%)	1 (7%)	47 (5%)	1 (13%)	41 (4%)	-
19 (2%)	0 (0%)	8 (0,8%)	0 (0%)	6 (0,6%)	0 (0%)
250 (26%)	6 (40%)	101 (10%)	0 (0%)	66 (7%)	0 (0%)

patients without PJI (OR 1.37 (0.39-4.87). Of the 18420 days of app use, an alert was sent 2589 (14%) times to 498 patients. In total, 141 (6.6%) annotations could be obtained from the electronic patient files confirming that patients had contacted the hospital based on the sent alert. This led to a change of treatment in 61 (43%) patients as summarized in Appendix C. Of the 16 patients who developed a PJI, an alert was sent in the preceding period to six patients which resulted in earlier outpatient evaluation or hospital admission in three patients.

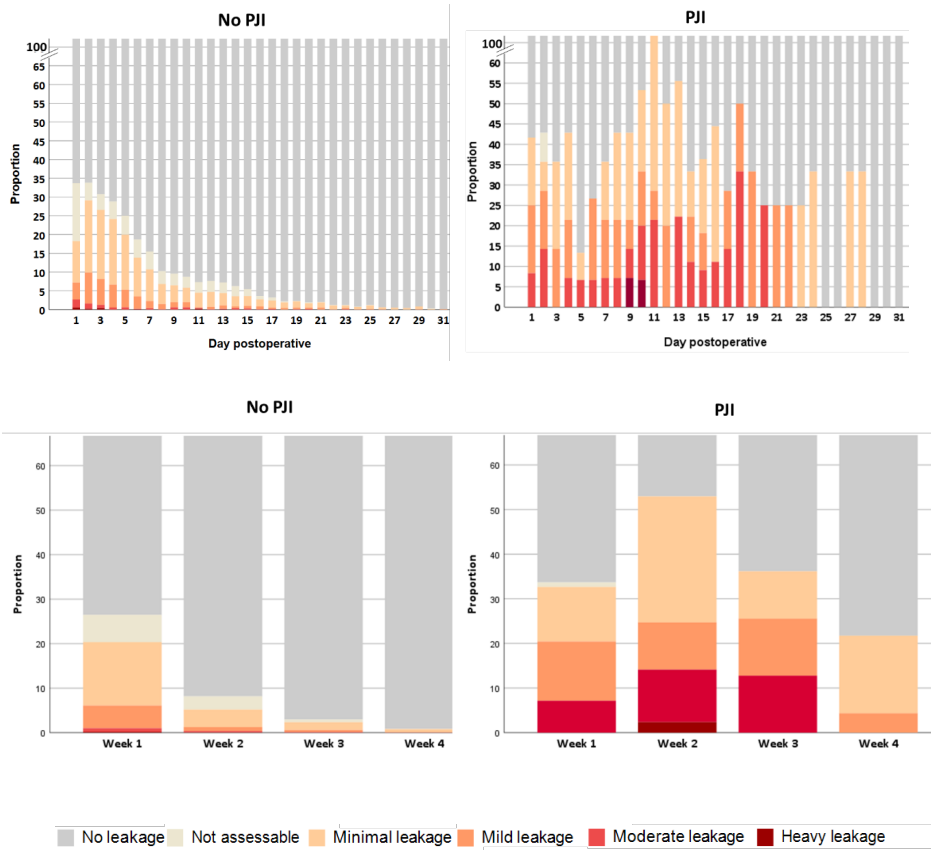


Figure 2. Reported extent and duration of postoperative wound drainage in patients with and without PJI.

Discussion

Principal findings

In the current study, a detailed overview of self-reported wound characteristics in the first month after arthroplasty while using a mobile wound care app provided important clinical insights. Complete absence of wound drainage during the first postoperative month was a sensitive and specific predictor of recovery without PJI. From the second week onward, wound drainage was strongly associated with the occurrence of PJI, but the positive predictive value remained low. Generation of an alert by the algorithm did not adequately identify patients with PJI.

Strengths and weaknesses

A major strength of this study is the unbiased prospective and daily information of exactly defined postoperative wound characteristics as provided by patients with an easy-to-

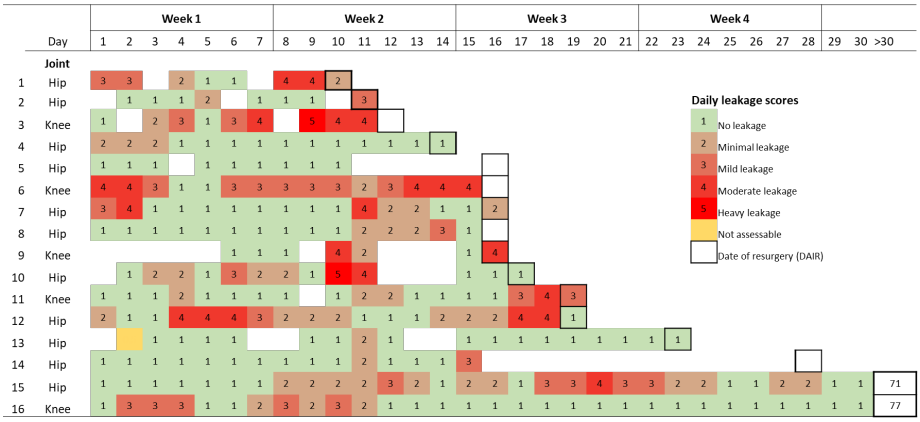
use smartphone application. Another strength is the large number of included patients without PJI that enabled us to create infographics of uncomplicated wound drainage for several subgroups of patients (Table 4).

Table 4. Percentage of postoperative wound drainage in 1003 patients without PJI during the postoperative course.

	Any wound leakage				Mild to heavy wound leakage				
	Hip		Knee		Hip		Knee		
	BMI < 30	BMI ≥ 30	BMI < 30	BMI ≥ 30	BMI < 30	BMI ≥ 30	BMI < 30	BMI ≥ 30	
DM	22%	30%	25%	72%	0%	10%	15%	38%	Week 1
No DM	40%	47%	62%	65%	12%	18%	27%	33%	
DM	11%	10%	19%	24%	0%	0%	10%	10%	Week 2
No DM	11%	16%	9%	9%	4%	8%	6%	4%	
DM	11%	0%	9%	24%	0%	0%	0%	14%	Week 3
No DM	5%	9%	7%	11%	1%	3%	3%	5%	
DM	0%	10%	0%	7%	0%	0%	0%	7%	Week 4
No DM	2%	2%	3%	6%	0%	2%	2%	1%	

Legend. BMI, Body Mass Index; DM, diabetes mellitus

Table 5. Drainage scores in 16 patients who developed a prosthetic joint infection



On verification in the patient files, *Patient 5* appeared to have stopped using the app on day 11 and subsequently developed wound drainage, redness and pain in the next three days without recording it in the app. On day 15, this patient was admitted with PJI. For *Patient 14*, no drainage data between day 14 and 28 could be retrieved from the patient notes.

This study has several limitations. The COVID-19 pandemic led to a temporary suspension of inclusions between March and May 2020 and continued to have a huge impact on the number of inclusions in the following year. The mean age of study participants (62.7 years for THA and 64.6 years for TKA) in our cohort was lower than the reported mean age in the Dutch Arthroplasty Registry involving all arthroplasties in The Netherlands (69.9 years for THA and 68.4 years for TKA in 2021), indicating that elderly may have been less willing to use the app. The relatively low number of patients with PJI in the study may have had an impact on the outcome. An even larger study would increase the precision of the results. Remarkably, patients who developed a PJI reported a relatively low proportion of wound redness and fever. We hypothesize that some patients with PJI symptoms may have visited their orthopedic surgeon without registering their symptoms in the app, but this remains speculative.

Table 6. Comparison of risk factors for failure in patients with and without PJI

	No PJI (n=1003)	PJI (n=16)	OR (95% CI)	Sens (%)	Spec (%)	PPV (%)	NPV (%)
Any drainage							
1st week	489/978	10/16	1.67 (0.60-4.62)	63	50	2	99
2nd week	115/950	14/16	50.83 (11.41-226.51)	88	88	11	100
3rd week	76/999	7/11	21.25 (6.09-74.22)	64	92	8	100
4th week	25/999	1/4	12.99 (1.31-129,24)	25	97	4	100
Moderate-heavy drainage							
1st week	47/978	4/16	6.60 (2.05-21.25)	25	95	8	99
2nd week	11/950	6/16	51.22 (15.84-165.65)	38	99	35	99
3rd week	1/999	5/11	103.23 (26.08-408.57)	45	100	83	99
4th week	1/999	0/4	-	0	100	0	100
New drainage after first week without drainage							
2nd week	28/480	5/6	80.71 (9.12-714.52)	83	94	15	100
3rd week	25/512	3/5	29.22 (4.67-182.85)	60	95	11	100
4th week	4/512	1/3	63.50 (4.74-850.04)	33	99	20	100
>5 cumulative leaking days during day 1-21							
2nd - 4th week	123/1003	9/16	9.20 (3.37-25.14)	56	88	7	99
Moderate to heavy drainage and/or fever and/or redness							
1st week	164/978	6/16	2.98 (1.07-8.31)	38	83	4	99
2nd week	68/950	7/16	10.09 (3.65-27.92)	44	93	9	99
3rd week	47/999	6/11	21.00 (6.21-70.99)	55	95	11	99
4th week	23/999	1/4	14.15 (1.42—141.17)	25	98	4	100
>2 alerts based on algorithm*							
1st week	141/978	3/16	1.37 (0.39-4.87)	19	86	2	98
2nd week	128/950	1/16	0.43 (0.06-3.27)	6	87	1	98
3rd week	69/999	0/11	-	0	93	0	99
4th week	47/999	0/4	-	0	95	0	100

Legend: PJI, prosthetic joint infection; OR, odd ratio; sens, sensitivity; spec, specificity; PPV, positive predicted value; NPV, negative predicted value.

* Algorithm is defined in Appendix A.

The short follow up of at least three months is a limitation of this study, in which we focused on the relation between wound drainage and early postoperative PJI. For late acute hematogenous PJI, initial wound drainage is probably not relevant because bacteremia is mostly the source of PJI. However, some patients with a chronic PJI will have been missed in our study and these patient may have had prolonged initial wound drainage providing a route for Coagulase-negative staphylococci to reach the implant and cause late chronic PJI. This would have resulted in an even stronger reported association between wound drainage and PJI than reported in this study. This needs to be further investigated in a follow up study.

Implications of our findings

This study has three important implications. First, moderate to heavy wound drainage in the third week strongly predicted PJI with a number needed to operate to diagnose one PJI of 1.2 patients. Although this predictor was only derived from a small subset of patients with PJI, moderate to heavy drainage was nearly absent in patients without PJI. Therefore, these patients need urgent clinical assessment of the postoperative wound to decide whether the patient should be operated for a suspected PJI or not.

Second, persistent wound drainage and wound drainage in the second and third postoperative week was strongly associated with development of PJI. However, positive predicted values were low due to the many patients with wound drainage during those weeks who did not develop PJI. If all patients with any form of drainage during the second postoperative week were regarded as suspected PJI, ten patients would need to be operated to find one PJI. This indicates that, even with a strong association between drainage and PJI, wound drainage alone is not an accurate predictor for presence of PJI in this group. The strength of the association did not increase significantly when fever and wound redness were added to wound drainage as risk factors, which may relate to the earlier mentioned low proportion of these symptoms reported by patients.

Third, wound drainage in the first postoperative week was not indicative of PJI. The high proportion of reported wound drainage during this week (Table 3, Figure 4) is explained by several factors: (1) drainage was recorded from the very first day postoperative day (not from discharge from hospital), (2) minimal wound drainage could have occurred during only one day of this week to be counted as wound drainage and (3) drainage was minimal (defined as <2x2cm on the gauze) in 87% of the patients with drainage in the first week (424/489 patients). Only 5% (n=51) of patients in this group had moderate to heavy wound drainage. In the second postoperative week wound drainage dropped down to 12%, again with minimal drainage (<2x2cm on gauze) in most (85%) of these patients.

This study confirmed that in patients without any wound drainage, an early postoperative PJI is very unlikely. With mobile health applications, this subgroup of patients can be easily identified during follow up and fewer outpatient visits may be needed during follow up which may reduce costs. The postoperative use of bandages during the first weeks to cover the postoperative wound may have resulted in underreporting of wound drainage. However, the impact was estimated to be similar in patients with and without PJI as the use of bandages was identical for all patients. We also assessed whether the closing technique (use of either glue or staples) was associated with duration of postoperative wound drainage after hip arthroplasty during the first two weeks, which was not the case (staples 3.2 days, glue 2.9 days, $p = 0.52$).

Only one out of the 16 patients with a PJI received more than two alerts prior to the PJI, indicating that the used algorithm was inadequate for predicting PJI. This may be explained by the low threshold in the algorithm for sending alerts secondary to pain and mild wound drainage. Many alerts were sent for minimal wound drainage or relatively mild pain scores not related to PJI. Unfortunately, a low number of alert-based treatment adaptations could be retrieved from the patient files making evaluation of the alerts send by the application speculative. Patients apparently made the right decision not to call their physician as no PJI occurred in 98% of them. The predictive value of the algorithm may be improved by using a machine learning algorithm, making iterative changes when the number of data increases thus allowing an automated update of the algorithm. Adding parameters like an increase in C-reactive protein may also increase the yield of the algorithm. Further, based on the current study, no "at-risk" points should be given for minimal wound drainage and low pain scores.

Conclusions

Detailed knowledge of the extent and duration of wound drainage after arthroplasty is vital for orthopedic surgeons who consider to reoperate patients with postoperative wound drainage for a suspected PJI. In this study, in which a mobile health application was used to monitor patients after arthroplasty, PJI was very unlikely in patients without any wound drainage. From the second week onward, wound drainage was strongly associated with the occurrence of PJI, but the sensitivity and positive predictive value of wound drainage as a single predictor for PJI was low. Due to the limited follow up of three months, some patients with a late chronic PJI may have been missed. The insights from this study may help clinicians evaluate postoperative patients who present with a leaking wound. Future research should focus on optimizing the algorithm, thereby improving the predictive value of the alert function.

Author contributions

Conceptualization: HS, RN, LV, MB, CL. Formal analysis : HS, MB, RZ. Software: RZ. Funding acquisition: HS, MB, RN. Investigation: RM, BE, WZ, TG, JL, RW, RP, MS, PJ, KB. Original draft preparation: HS. Review and editing: HS, BE, RM, WZ, TG, JL, RW, RP, MS, PJ, KB, RN, LV, MB. Supervision: MB, LV, RN

Ethical statement

The study was conducted according to the principles of the Declaration of Helsinki. The study was approved by the ethics review committee of Leiden University Medical Center and a waiver was obtained to use electronic instead of written informed consent(P18.220). The use of the app for this study was approved by the Dutch Health Inspectorate (reference number VGR2O1 1434).

Acknowledgments

This trial was made possible by the collaborative efforts of doctors and nurses at all participating hospitals, mostly during the covid19 pandemic. We thank everyone who contributed their time and expertise, in particular the trial participants and local research coordinators. Their input and understanding were important in ensuring the success of this study.

Competing interests

The authors declare they have no conflicts of interest. This work was supported by an unrestricted grant from the Innovation Fund of Dutch Health Insurers (grant number 3687) and Foundation De Merel (grant number BS094 057).

**Wound care app study group:* Enrike van der Linden, Leiden University Medical Center; Demien Broekhuis, Leiden University Medical Center; Menno Benard and Astrid Mol, Alrijne Hospital; Marijn Rutgers, Jantsje Pasma, Roos Bazuin and Nina Mathijssen: Reinier Reinier Haga Orthopedic Center; Nienke Willigenburg en Myke van der Hoorn, Onze Lieve Vrouwe Gasthuis; Baukje Dijkstra, Medical Center Leeuwarden; Lex de Jong, Rijnstate Hospital; Liesbeth Paans and Wouter Bekkers, Elisabeth Tweestedenziekenhuis, Henry Haan and Martin Stevens, University Medical Center Groningen; Max Reijman, Erasmus Medical Center Rotterdam; Carolien Kooijman and Hendrike Hoogeboom, Nij Smellinghe Hospital; Chris Meijer and Hieron Maree, Park Medical Center, + all authors listed above this article

References

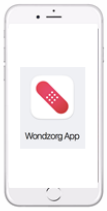
1. Hiligsmann M, Cooper C, Arden N, et al. Health economics in the field of osteoarthritis: an expert's consensus paper from the European Society for Clinical and Economic Aspects of Osteoporosis and Osteoarthritis (ESCEO). *Seminars in arthritis and rheumatism* 2013;43(3):303-13. doi: 10.1016/j.semarthrit.2013.07.003 [published Online First: 2013/09/03]
2. Kurtz SM, Lau E, Watson H, et al. Economic burden of periprosthetic joint infection in the United States. *J Arthroplasty* 2012;27(8 Suppl):61-5 e1. doi: 10.1016/j.arth.2012.02.022 [published Online First: 2012/05/05]
3. Zimmerli W, Trampuz A, Ochsner PE. Prosthetic-joint infections. *N Engl J Med* 2004;351(16):1645-54. doi: 351/16/1645 [pii];10.1056/NEJMra040181 [doi]
4. Wagenaar FBM, Lowik CAM, Zahar A, et al. Persistent Wound Drainage After Total Joint Arthroplasty: A Narrative Review. *J Arthroplasty* 2019;34(1):175-82. doi: 10.1016/j.arth.2018.08.034 [published Online First: 2018/09/25]
5. Patel VP, Walsh M, Sehgal B, et al. Factors associated with prolonged wound drainage after primary total hip and knee arthroplasty. *J Bone Joint Surg Am* 2007;89(1):33-8. doi: 10.2106/JBJS.F.00163 [published Online First: 2007/01/04]
6. Almeida RP, Mokete L, Sikhauli N, et al. The draining surgical wound post total hip and knee arthroplasty: what are my options? A narrative review. *EFORT Open Rev* 2021;6(10):872-80. doi: 10.1302/2058-5241.6.200054 [published Online First: 2021/11/12]
7. Weiss AP, Krackow KA. Persistent wound drainage after primary total knee arthroplasty. *J Arthroplasty* 1993;8(3):285-9. [published Online First: 1993/06/01]
8. Parvizi J, Gehrke T, Chen AF. Proceedings of the International Consensus on Periprosthetic Joint Infection. *Bone Joint J* 2013;95-B(11):1450-52. doi: 95-B/11/1450 [pii];10.1302/0301-620X.95B11.33135 [doi]
9. McNally M, Sousa R, Wouthuyzen-Bakker M, et al. The EBJIS definition of periprosthetic joint infection. *Bone Joint J* 2021;103-B(1):18-25. doi: 10.1302/0301-620X.103B1.BJJ-2020-1381.R1 [published Online First: 2021/01/01]
10. Sanger PC, Hartzler A, Han SM, et al. Patient perspectives on post-discharge surgical site infections: towards a patient-centered mobile health solution. *PLoS One* 2014;9(12):e114016. doi: 10.1371/journal.pone.0114016 [published Online First: 2014/12/02]
11. Gray RT, Sut MK, Badger SA, et al. Post-operative telephone review is cost-effective and acceptable to patients. *Ulster Med J* 2010;79(2):76-9. [published Online First: 2010/12/01]
12. Chua ME, Saunders MA, Bowlin PR, et al. Impact of smartphone digital photography, email, and media communication on emergency room visits post-hypospadias repair. *Can Urol Assoc J* 2017;11(3-4):E134-E37. doi: 10.5489/cuaj.4170 [published Online First: 2017/05/19]
13. Armstrong KA, Coyte PC, Brown M, et al. Effect of Home Monitoring via Mobile App on the Number of In-Person Visits Following Ambulatory Surgery: A Randomized Clinical Trial. *JAMA Surg* 2017;152(7):622-27. doi: 10.1001/jamasurg.2017.0111 [published Online First: 2017/03/23]
14. Scheper H, Derogee R, Mahdad R, et al. A mobile app for postoperative wound care after arthroplasty: Ease of use and perceived usefulness. *Int J Med Inform* 2019;129:75-80. doi: 10.1016/j.ijmedinf.2019.05.010 [published Online First: 2019/08/25]

15. Parvizi J, Gehrke T, Chen AF. Proceedings of the International Consensus on Periprosthetic Joint Infection. *Bone Joint J* 2013;95-B(11):1450-2. doi: 10.1302/0301-620X.95B11.33135 [published Online First: 2013/10/24]
16. Lowik CAM, Wagenaar FC, van der Weegen W, et al. LEAK study: design of a nationwide randomised controlled trial to find the best way to treat wound leakage after primary hip and knee arthroplasty. *BMJ open* 2017;7(12):e018673. doi: 10.1136/bmjopen-2017-018673 [published Online First: 2017/12/31]

Appendices

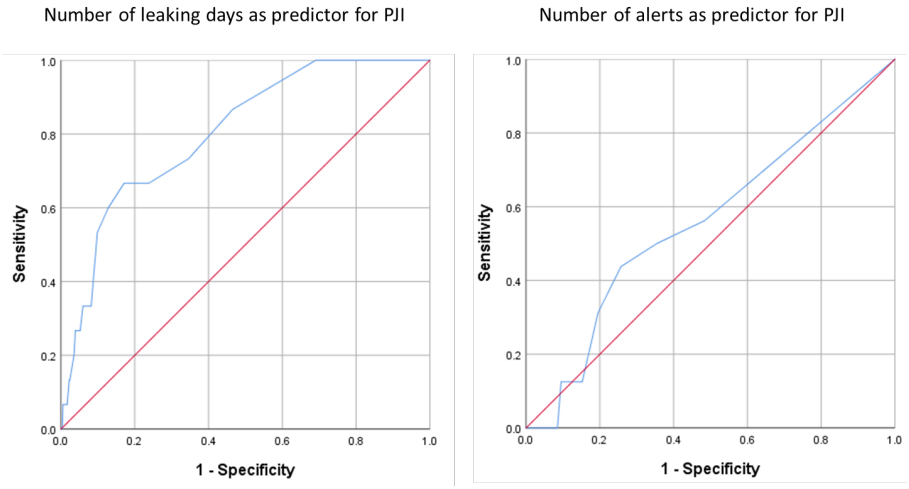
Appendix A. Calculated scores from the app and algorithm for sending alert to patients

Daily review	Answers	Points
1. Fever?	T <38^{o}C	0
	T 38-38.5^{o}C	2
	T 38-38.5^{o}C (>2days)	5
	T >38.5^{o}C	5
2. Wound leakage?	No	0
	Minimal(<2x2cm on bandage)	1
	Mild (>2x2cm on bandage)	2
	Moderate (1-2 bandages exchanged)	3
	Severe (>2 bandages exchanged)	4
	Not judgeable (e.g. plaster/dressing)	0
3. Redness of wound?	No/unchanged	0
	Increased redness compared to yesterday	2
4. VAS score	VAS ≤ 5	0
	VAS >3 (>3days)	3
	VAS >2pts increase in 1 day	3
	VAS 6 or 7	3
	VAS >7	4
	Total amount of points:	☐



Score is calculated daily. Alert sent if: ≥ 5 points *or*
≥ 4 points 2 consecutive days *or*
≥ 3 points 3 consecutive days

Appendix B. ROC using duration of leakage days or number of sent alerts as cutoff value for detecting PJI.



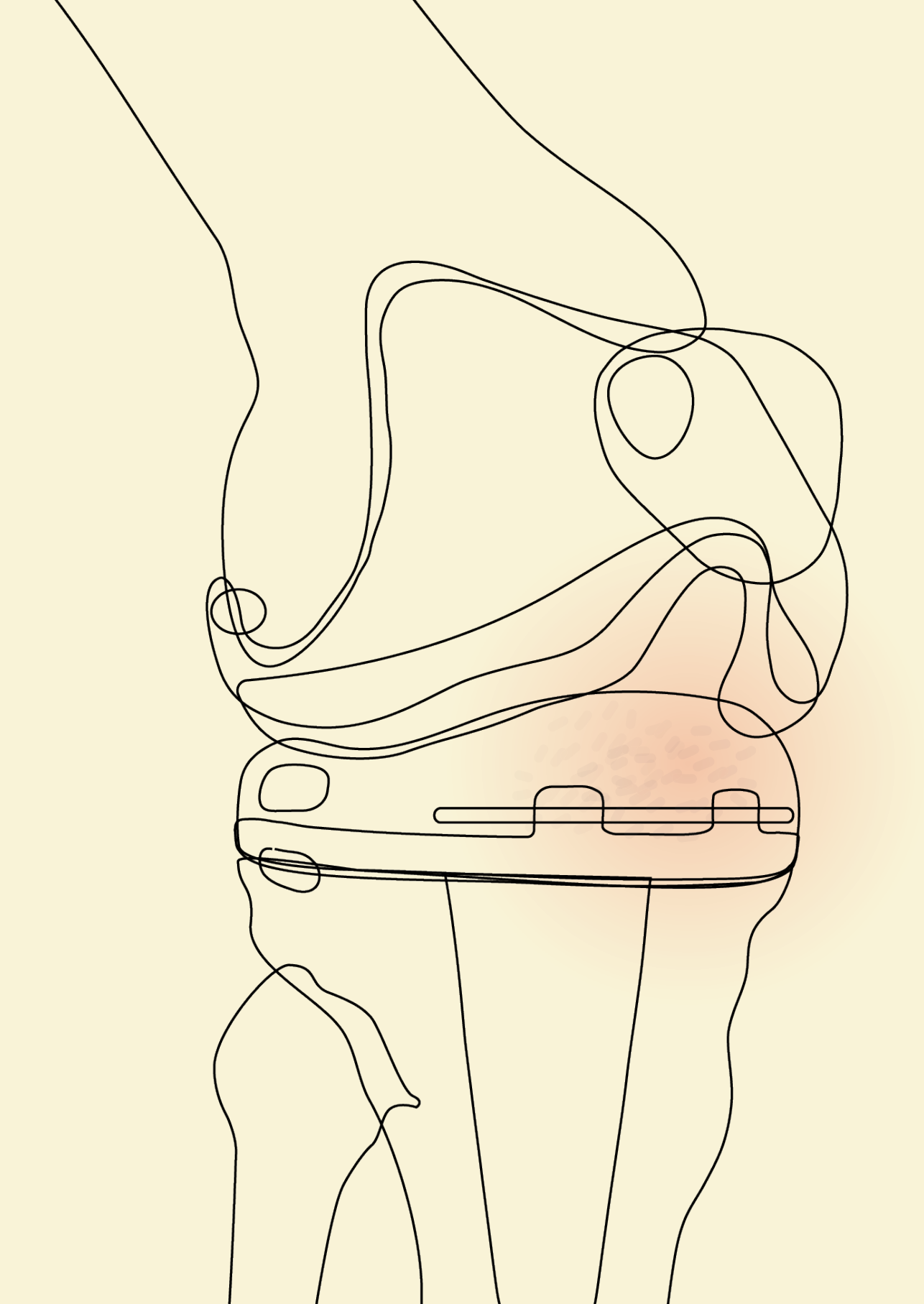
Appendix C. Actions taken on algorithm-based alerts generated by the app from 18437 daily reports

	All	No PJI (n=1013)	PJI (n=16)
Total alert count	2590 (14%)		
Alerts received (n patients)	498/1019 (48.9%)		
Alert 1st week postoperative (n patients)	423/498 (84.9%)		
Alert 2nd week postoperative (n patients)	232/498 (46.6%)		
Alert 3rd week postoperative (n patients)	97/498 (19.5%)		
Alert 4thweek postoperative (n patients)	68/498 (7.0%)		
Alerts per individual patient (median)	3		
Reported patient-physician contact based on alerts	141/2124 (6.6%) *	135 (13%)	6 (40%)
Outcome of patient-physician contact			
No action needed	51 (36%)	51	0
Adjust pain medication	34 (24%)	34	0
Earlier outpatient evaluation	24 (17%)	22	2
Admission to hospital	3 (2%)	2	1
Other	29 (21%)	26	3

* From 466 alerts, patient files could not be checked for placed phone calls

^

$ Practical wound management advice, Deep Venous Thrombosis excluded, patient not yet discharged



Part II. Evaluation of current antimicrobial strategies for PJI

CHAPTER 4

Outcome of debridement, antibiotics and implant retention for staphylococcal hip and knee prosthetic joint infections, focused on rifampicin use: a systematic review and meta-analysis.

Henk Scheper, Laura Maxime Gerritsen, Bart G. Pijls, Suzanne A. van Asten, Leo G. Visser, Mark G.J. de Boer

Open Forum Infect Dis 2021 Jul 1;8(7)

Abstract

The treatment of staphylococcal prosthetic joint infection (PJI) with debridement, antibiotics and retention of the implant (DAIR) often results in failure. An important evidence gap concerns the treatment with rifampicin for PJI. A systematic review and meta-analysis were conducted to assess the outcome of staphylococcal hip and/or knee PJI after DAIR, focused on the role of rifampicin. Studies published until September 2nd, 2020 were included. Success rates were stratified for type of joint and type of micro-organism. Sixty-four studies were included. The pooled risk ratio for rifampicin effectiveness was 1.10 (95% CI 1.00-1.22). Pooled success rate was 69% for S. aureus hip PJI, 54% for S. aureus knee PJI, 83% for CNS hip PJI and 73% for CNS knee PJI. Success rates for MRSA PJI (58%) were similar to MSSA PJI (60%). The meta-analysis indicates that rifampicin may only prevent a small fraction of all treatment failures.

Introduction

A prosthetic joint infection (PJI) is a severe complication of orthopedic surgery and associated with significant morbidity and mortality. *Staphylococcus aureus* (*S. aureus*) or Coagulase-negative staphylococci (CNS) are the most common causative pathogens of PJI, accounting for about two-third of all cases[1]. Treatment of acute PJI, aimed at maintaining the implant, consists of thorough surgical debridement of the implant and of the infected tissue around the implant, followed by antibiotic treatment (summarised as DAIR: Debridement, Antibiotics and Implant Retention). Nevertheless, failure rates with this treatment strategy are high, ranging from 10% to 45% in some of the largest studies[2, 3]. An important evidence gap concerns the causes for these high failure rates. The type of joint, the type of micro-organism and the antibiotic treatment that was used for PJI are risk factors that have been put forward to explain these high failure rates. Most international guidelines have adopted rifampicin combination therapy as the cornerstone antibiotic treatment for staphylococcal PJI treated with DAIR, based on experimental animal models, one randomised trial and several cohort studies. However, rifampicin combination therapy is associated with significant side effects and drug-drug interactions, making its use less patient-friendly [4, 5]. Moreover, the literature regarding the effect of rifampicin combination therapy against staphylococcal hip and knee PJI after DAIR has not yet been explored systematically. Most observational PJI studies also included patients with PJI caused by other micro-organisms. Furthermore, not all studies specify details regarding the outcome per affected joint (hip or knee) or per causative staphylococcal species (*S. aureus* or CNS), both of which may influence success rate. Therefore, we conducted a literature search to systematize and appraise the available evidence concerning outcome of staphylococcal PJI treated with DAIR, with a specific focus on the outcome with or without rifampicin use. A secondary objective was to relate outcomes to the type of joint (hip or knee), the type of micro-organism (*S. aureus* and CNS) and susceptibility to methicillin (methicillin-resistant *S. aureus*, MRSA and methicillin-sensitive *S. aureus*, MSSA).

Methods

Search strategy and selection criteria

The reporting of this systematic review and meta-analysis is in accordance with the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) statement. The population of interest included all patients evaluating the outcome after DAIR for the treatment of staphylococcal hip and/or knee PJI, as defined by IDSA or MSIS criteria[6]. Studies that also included other types of surgical strategy, other joints or

other micro-organisms were only included if the outcome was quantified separately for the variables of our interest. The following exclusion criteria were applied: Studies that included patients with superficial wound infection, case reports and studies reporting 20 patients or less with staphylococcal PJI[7]. A meta-analysis was performed for the studies in which patients treated with rifampicin could be compared with patients not treated with rifampicin. The search was limited to articles published until September 2nd, 2020. Articles were identified searching PubMed, Cochrane Library and Embase databases (Supplemental Table 1). In addition, bibliographies of relevant articles were cross-checked for references missing in the original search. Two independent reviewers (H.S. and L.M.G.) reviewed all studies. A third reviewer (MdB) was consulted if disagreements between reviewers could not be solved.

Table 1. Reported outcome after DAIR, stratified for micro-organism and/or type of joint using individual patient data from 64 included studies

Micro-organism and/or type of joint	n studies*	n patients*	pooled success rate of all individual patient data	RR (95%CI)#
All	64	4380	60%	-
Per micro-organism				
S. aureus	54	2922	61%	ref.
CNS	36	761	74%	1.50 (1.32-1.70)
Per affected joint				
Knee	27	1106	55%	ref.
Hip	24	904	69%	1.45 (1.29-1.63)
Per affected joint *and* micro-organism				
S. aureus knee PJI	19	692	54%	ref.
CNS knee PJI	12	187	73%	1.72 (1.33-2.21)
S. aureus hip PJI	19	547	69%	1.48 (1.27-1.72)
CNS hip PJI	13	145	83%	2.66 (1.85-3.84)

* The columns 'n studies' and 'n patients' displays the number of studies and patients for which the specific outcome regarding affected joint and/or micro-organism was reported. For example: one study could report outcome for both *S. aureus* and CNS but not stratifying outcome for type of joint, while other studies only reported outcome for the total population without stratification for either type of joint or micro-organism. Therefore, numbers in this table cannot be summed.

Relative Risks for success were calculated for micro-organisms (with *S. aureus* PJI as reference), for type of joint (with knee PJI as a reference) and for the 4 groups (with *S. aureus* knee PJI as a reference).

Data analysis

Texts of selected abstracts were reviewed, as were article texts of abstracts that could not be excluded based on abstract review alone. Data from each study were entered in an SPSS database. Information extracted included study design, number of patients with *S. aureus* and/or CNS PJI, number of hip and/or knee PJI, year of publication, duration of follow-up, rifampicin use (number of patients receiving rifampicin) and treatment outcomes for all these subcategories. As there is no universally accepted definition for treatment success or failure after PJI, the definitions used by the included paper were used. We contacted study authors and requested individual patient-level data if rifampicin data were not clearly specified.

Assessment of quality of evidence

Estimates of associations in observational studies may deviate from true underlying relationships due to confounding or biases. Confounding may occur as patients with comorbidity or use of immunosuppressants, implying a higher a priori risk for a poor outcome, may not be selected for rifampicin treatment. Survival bias occurs when only patients 'surviving' the first weeks after debridement are included in the rifampicin group. The Newcastle-Ottawa Quality Scale was used to assess the quality of the studies included in the meta-analysis (Supplemental table 3). As this scale only addresses basic methodological factors and not important confounding factors or survival bias, studies will also be reviewed qualitatively in the discussion.

Statistical methods

For the meta-analysis, we used the Hedges random-effects model to pool the risk ratio (RR) of individual studies in order to estimate an overall RR along with its associated confidence interval (CI). The choice for a random effects method was based on the assumption that underlying risk factors for outcome were expected to vary between studies regarding underlying host comorbidities, type of joint and the severity of PJI. Patients were excluded from the meta-analysis if failure occurred in the first week after debridement and before initiation of rifampicin, to prevent survivor bias. The extent of statistical heterogeneity was assessed by calculating I^2 statistics. A funnel plot was constructed for studies reporting the primary outcome to assess the possibility of publication bias. Success rates were compared in predetermined subgroups (hip versus knee, *S. aureus* versus CNS, MRSA versus MSSA) using *t test*. A linear regression model, including success rate, proportion of rifampicin use and type of joint was used to further explore the relationship between rifampicin use and success rates. Descriptive statistics were performed using SPSS 23.0 (IBM Corp., Amonk, NY: USA). Stata was used for the meta-analysis (StataCorp, version 16, Texas, USA). The study-protocol was registered a-priori with PROSPERO (registration number CRD42020155132).

Results

Study selection and study characteristics

The review process identified 2186 articles, of which 263 full text articles were assessed for eligibility (Figure 1). In total, 64 studies (4380 patients) were included, published between 1990 and September 2nd, 2020 (Supplemental Table 2). Only two studies were published before 2005. All studies were observational cohort studies (3 prospective, 59 retrospective), except for two randomized controlled trials. The median study size was 50 patients; ten studies included more than 100 patients. *S. aureus* was the causative micro-organism in 3142 patients, CNS in 915 patients and the staphylococcal species was not specified in 323 patients. Of 1797 patients with *S. aureus* PJI in which the methicillin susceptibility of the isolates was reported, 416 (21%) were MRSA. Use of rifampicin for staphylococcal PJI was mentioned in 49 studies. Of those studies, outcome of treatment with or without rifampicin was reported in 30 studies (Table 2, also Supplemental Table 4). Except for one RCT, no studies compared baseline characteristics between patients treated and not treated with rifampicin. The study by Karlsen and colleagues was the only randomised controlled trial that could be included in the meta-analysis. In this study, 48 patients with staphylococcal PJI were randomised between rifampicin combination therapy (23 patients) and beta-lactam monotherapy (25 patients).

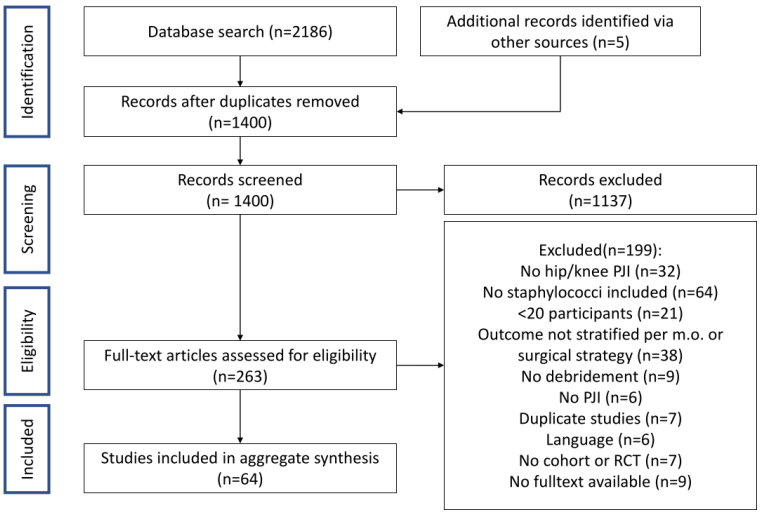


Figure 1. Flow chart of study selection.

Outcome after DAIR related to micro-organism

Outcome of treatment for staphylococcal PJI is presented in table 1. The pooled success rate in all included studies was 60%. In smaller cohorts (<100 patients), the reported success rates varied from 23% to 90% (figure 4). Cure rates in the two largest cohort

studies (both containing more than 300 patients and likely more closely reflecting a real-life clinical situation) were 54% and 56%[2, 8]. Pooled success rate for *S. aureus* PJI after DAIR was 62% (2922 analyzed patients in 54 studies) and for CNS PJI 73% (36 studies, 760 patients; table 1). Outcome for MRSA and MSSA PJI was reported in 25 and 28 studies, respectively (table 3); success rate after DAIR was not different between both groups (MRSA 58%, MSSA 60%, p=0.459). Outcomes between MRSA and MSSA PJI were not different when stratified for type of joint (data not shown). Pooled success rate of *S. aureus* PJI after DAIR was 67% if PJI occurred within 3 months after arthroplasty and 49% in patients with later onset of *S. aureus* PJI (990 analysed patients in 9 studies)[2, 9-16] .

Outcome after DAIR related to type of joint

Outcome per affected joint was specified in 33 studies. Pooled success rate after DAIR for *S. aureus* hip PJI was 69%, while pooled success rate after *S. aureus* knee PJI was 54% (table 1). Pooled success rates after DAIR for CNS hip PJI was 83% and 73% for CNS knee PJI. Using linear regression analysis, reported success rates positively correlated with the proportion of included hip PJI per study: success rates increased from 54% in studies with <25% of patients with hip PJI to 82% in studies with >75% of patients with hip PJI (p=0.002), indicating that reported outcome of PJI is strongly affected by the type of joint included in studies (figure 5). The high success rates for hip PJI could not be attributed to rifampicin use: success rates were 83% for patients on rifampicin and 82% for patients who were not treated with rifampicin (RR 1.01, 95% CI 0.85-1.20; evaluable in four studies with 157 patients, table 2).

Table 2. Outcome of 30 studies that reported individual patient data regarding the use of rifampicin or not.

	N studies	N patients	Cure with rifampicin#	Cure without rifampicin#	RR (95%CI)
Hip PJI*					
S. aureus	0				
CNS	0				
Combined	4	157	102/123 (83%)	28/34 (82%)	1.01 (0.85-1.20)
Knee PJI*					
S. aureus	1	22		9/22 (41%)	
CNS	0				
Combined	2	108	56/69 (81%)	17/34 (50%)	1.62 (1.14-2.31)
Hip and knee PJI$					
S. aureus	3	135	100/125(80%)	4/10 (40%)	2.00 (0.93-4.29)
CNS	0				
Combined	24	1652	903/1298 (70%)	186/354 (53%)	1.32 (1.19-1.47)

#Pooled individual patient data in N studies

*Per category studies are included if outcome is reported apart for *S. aureus* and/or CNS apart or combined if outcome for all staphylococci is summarized

$Studies are included in this category if outcome was reported only for hip and knee PJI together

Table 3. Outcome of MSSA versus MRSA PJI treated with DAIR

Study	N studies*	N patients	Pooled success rate#
MSSA PJI	28	1381	60%
MRSA PJI	26	416	58%
Hip MSSA PJI	2	32	81%
Hip MRSA PJI	1	12	92%
Knee MSSA PJI	3	56	66%
Knee MRSA PJI	3	78	64%

*Per category, studies were included if they reported specific or combined outcome for hip and/or knee MSSA and MRSA.
based on individual patient data
MSSA: methicillin-susceptible *Staphylococcus aureus*, MRSA: methicillin-resistant *Staphylococcus aureus*

Outcome after DAIR related to treatment with rifampicin

The reported success rates over the years, stratified by treatment with rifampicin, are shown in figure 2. Success rates were higher in studies in which rifampicin was prescribed (64% in 34 studies with 2884 patients) compared to studies in which rifampicin was not prescribed or not mentioned by the authors (44% in 18 studies with 976 patients). In twelve studies, all included patients were treated with rifampicin resulting in a pooled success rate of 71% (table 2). These studies were likely hampered by selection bias because outcome of patients who did not use rifampicin were not evaluated herein. Twelve observational studies and one randomized controlled trial reported outcome for both patients treated and not treated with rifampicin. In two of these studies, the group of patients without rifampicin was too small for comparative evaluation[17, 18]. Outcome of the remaining 11 studies was evaluated with a random effects meta-analysis (figure 3). Survivor bias could be corrected in two of those studies, in which 5 out of 17 and 6 out of 13 patients failed before initiation of rifampicin[19, 20]. From one study, comparing two historical groups and one prospective group, only the historical groups were included in the meta-analysis because these groups could be compared with each other while a control group for the prospective cohort was absent. The only included RCT in the meta-analysis (by Karlsen and colleagues) reported similar cure rates between the rifampicin group (74%) and the beta-lactam group (72%)[21]. The pooled risk ratio for rifampicin effectivity from 11 studies in the meta-analysis was 1.10 (95% CI 1.00-1.22). The funnel plot was asymmetric (Supplemental Figure 1). A trim-and-fill analysis to explore this possible publication bias suggested four missing studies, which after correction would result in an adjusted relative risk for success of 1.04 (95%CI 0.94 tot 1.14).

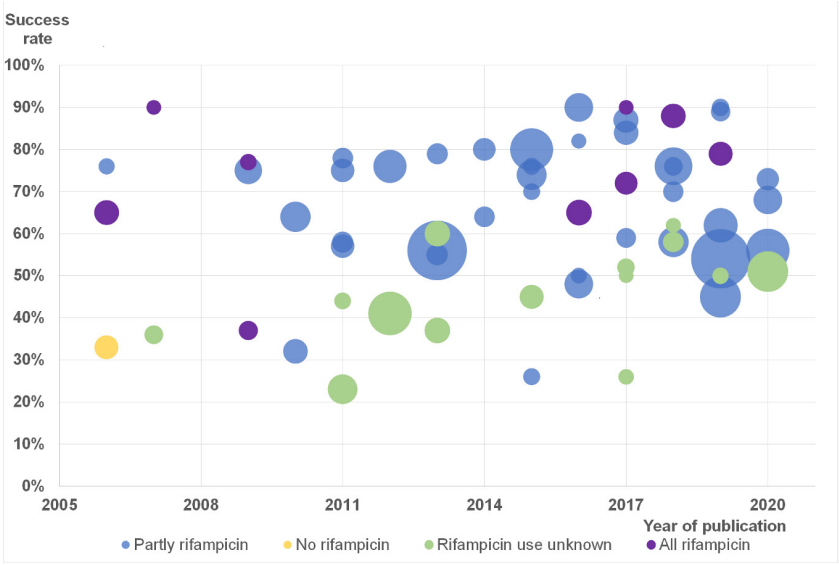


Figure 2. Success rates over the years for staphylococcal PJI treated with DAIR and related to use of rifampicin. Different bubble sizes represent differences in study size

Discussion

Despite gradually improving success rates over the years, the reported outcome of staphylococcal PJI is still heterogeneous, ranging from 23% to 90%. Overall, the pooled risk ratio for success was slightly higher in patients treated with rifampicin. Success rates were considerably better for hip and CNS PJI than for knee and *S. aureus* PJI. Success rates of MRSA and MSSA PJI after DAIR were similar. Of note, the ratio of *S. aureus* to CNS PJI remained stable over the years (between 72-76%), indicating that success rates are probably not influenced by changing epidemiology of causative staphylococci.

The pooled estimated effect of rifampicin on treatment outcome in our meta-analysis differs from a recently published meta-analysis that did not find a positive association between treatment with rifampicin and success rates[22]. Several studies in that meta-analysis included other micro-organisms than staphylococci or patients with other surgical strategies whom we excluded[23-26]. Moreover, with a broader search strategy, we were able to include seven other studies in our meta-analysis.

Interpreting the association between rifampicin and success rates after DAIR in the meta-analysis is complicated by survival bias and selection bias, as ten studies in the

meta-analysis were observational. In three studies, survival bias could be ruled out by excluding patients who failed early after debridement and before start of rifampicin, [2, 19, 20] but survival bias was likely present in more studies. The positive association between duration of rifampicin and success rates after DAIR in the study of Becker and colleagues could be explained by survival bias and selectively excluding patients from the analysis who developed a failure while on rifampicin treatment[27, 28]. Lora-Tamayo and colleagues described the strongest association between rifampicin use and outcome. This study addressed survivor bias and performed multivariate regression analysis to correct for confounding factors, which did not change the outcome of the study[2]. The trim- and fill- analysis suggested that publication bias have influenced the outcome of the meta-analysis. However, this analysis is a statistical measure that presumes that negative studies were not published, which in our opinion is not very likely given the many studies presented in this review with negative results.

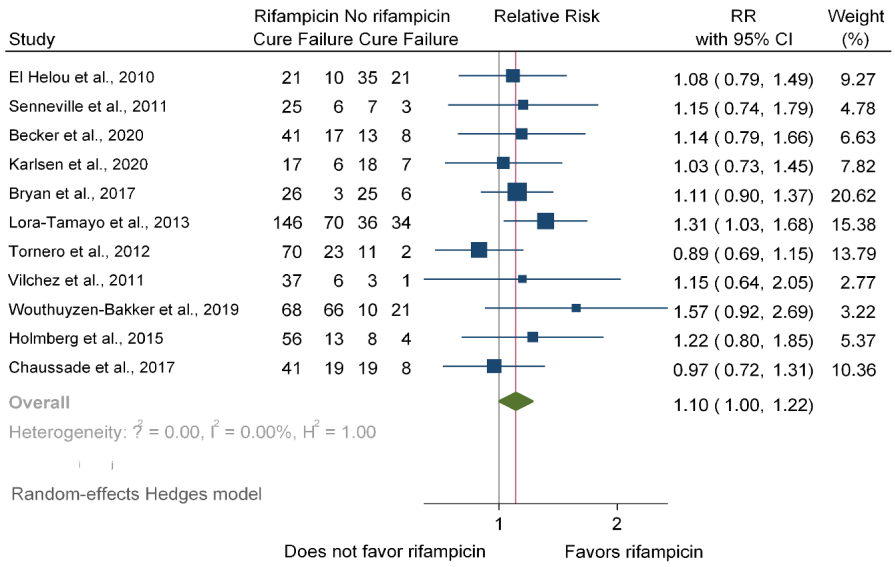


Figure 3. Meta-analysis of 11 studies in which outcome for staphylococcal PJI after DAIR could be compared between patients treated and not treated with rifampicin

The point estimate (relative risk, RR) for each study is represented by a square. The 95% CI for each study is represented by a horizontal line intersecting the square. The size of the square represents the relative precision of the study estimates: the bigger the square the more precise the study was.

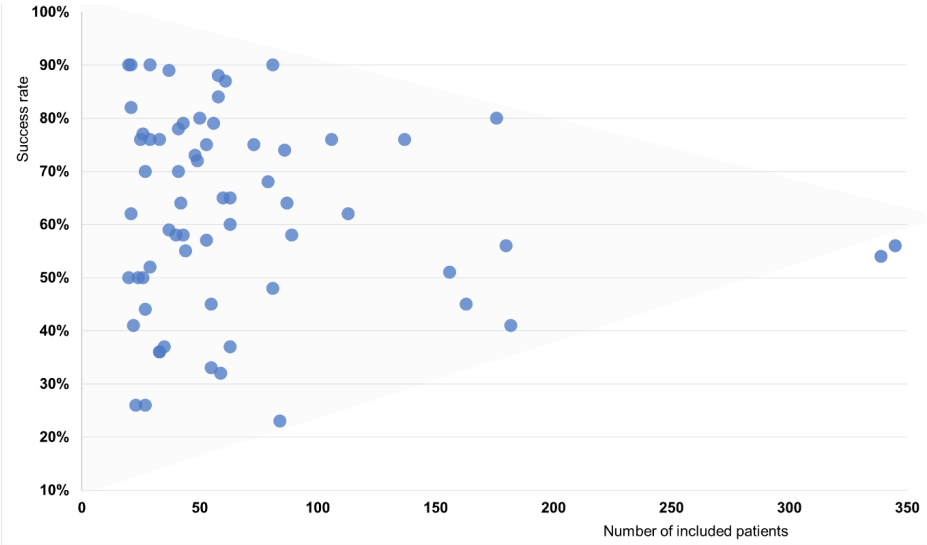


Figure 4. Relation between study size and outcome of staphylococcal PJI treated with DAIR (n = 64 studies)

As most studies in this review were observational, confounding factors that influence both the choice for antibiotic strategy and outcome after DAIR were present in these studies. Unfortunately, a comparison of baseline characteristics between rifampicin and non-rifampicin users is nearly absent in the literature summarised in this review. Survival bias may explain the increased effectiveness of long-term rifampicin compared to short-term rifampicin in the study of Lesens and colleagues, because these patients were only analysed in the group with long-term rifampicin if experience treatment failure during the first weeks of treatment[11]. Confounding by indication was described in the studies of Morata and colleagues and Ascione and colleagues in which patients who were not treated with rifampicin had diabetes, rheumatoid arthritis and liver disease more often[23, 26].

The well-known RCT of Zimmerli and colleagues (1998) was excluded from this review due to the low number of patients (18 patients with PJI of whom eight received rifampicin) and because outcome was not stratified per micro-organism (both *S. aureus* and CNS included) and type of infection (both osteosynthesis-associated infection and PJI were included). Patients were randomised in this trial between rifampicin combination therapy or ciprofloxacin monotherapy[29]. Intention-to-treat analysis showed a nonsignificant 89% versus 60% cure rate in favour of rifampicin; significance was reached in the per protocol analysis. However, the choice for ciprofloxacin monotherapy in the control arm, nowadays regarded as inferior therapy for staphylococcal PJI, played a major role in the outcome as

four out of five failures this group were due to ciprofloxacin resistance. The RCT of Karlsen and colleagues contained three times as much patients than the trial of Zimmerli and colleagues and had a different comparator arm (beta-lactams instead of ciprofloxacin)[21] .

The timing of rifampicin initiation and the duration of treatment with rifampicin may also affect outcome. In the two randomized controlled trials discussed above and one observational study, rifampicin was started immediately or from day one postoperatively[21, 29, 30]. In these studies, rifampicin resistance had not developed in patients with positive cultures after failure. Rifampicin resistance in patients with failure after DAIR has been reported, but this was in patients who were not treated with adequate debridement or with combination therapy[31, 32]. Whether the duration of rifampicin combination therapy affects outcome is not sure. Treatment duration was three months in most studies included in this review. In some observational studies, shorter rifampicin treatment was associated with more treatment failure, but these results should be interpreted cautiously as studying treatment duration in observational studies is inherently affected by selection bias and survival bias[11, 27, 28]. More research is needed to gain more evidence regarding the timing and duration of rifampicin.

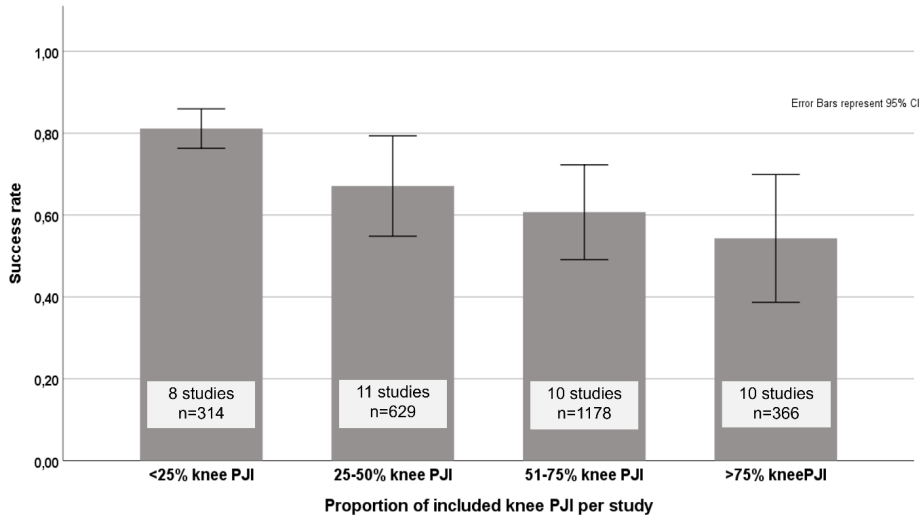


Figure 5. Success rates in 39 studies that could be categorized by knee-to-hip-ratio

This review reveals that success rates are strongly influenced by the ratio of knee-to-hip PJI per study. Are higher success rates, usually attributed to rifampicin use, in fact explained by a decreased knee-to-hip ratio in studies? To explore this further, we related the knee-to-hip ratio to rifampicin use. We unexpectedly found that the knee-to-hip ratio per study was inversely related to rifampicin use. The knee-to-hip PJI ratio in studies was 0.90 (meaning more knees than hips) if rifampicin was not used, 0.77 if rifampicin use was not mentioned, 0.40 if a certain proportion of patients used rifampicin and 0.35 in studies in which all patients were treated with rifampicin. As a derived measure we performed linear regression analysis with proportion of rifampicin use per study as predictor variable for success weighted by proportion of included knee PJIs. Results revealed that the significant correlation between rifampicin use and successful outcome (p=0.01) disappeared after correction for type of joint (p=0.17), indicating that both rifampicin and type of joint influence the outcome of PJI. We hypothesize that adjunctive rifampicin use will not yield a further increase in success rate in patients with hip PJI with a priori higher chances for cure. The poor outcomes of knee PJI may relate to the surgical debridement, which is more complicated for infected knee prostheses than for hip prostheses due the anatomical barriers that hinder a proper debridement of a knee prosthesis. Of note, outcome for knee PJI was better in patients treated with rifampicin (81%) compared to patients not treated without rifampicin (50%) (RR 1.62 (1.14-2.31), but this risk ratio could be obtained from only two studies.

The definition of treatment failure varied across included studies. In most studies a second debridement within the first three weeks of antibiotic treatment was not regarded as failure, while other studies defined all subsequent debridements as failure. Further, the use of chronic suppressive antibiotic therapy with a well-functioning prothesis is defined as a failure in some but not all studies, also affecting cure rates in studies[33]. Of all included studies, 30 studies did not report whether chronic antibiotic suppression was part of the definition of failure or was regarded as success in patients with a functioning prosthesis. Of note, success rates ware comparable between 30 studies that defined chronic suppressive antibiotics as failure (61%) and 34 studies that did not mention suppressive therapy or regarded suppressive therapy as success (60%), but interpretation is difficult as most studies did not specify the number of patients on suppressive antibiotic treatment. Uniform definitions of treatment failure are needed making comparison between studies more accurate.

In this review, higher success rates were reached in early postoperative PJI (within 3 months after arthroplasty) compared to later onset of PJI. Wouthuyzen-Bakker and colleagues reported lower treatment success of late acute (hematogenous) PJI compared to early postoperative PJI for both *S. aureus* (34% versus 75%) and CNS (46% versus 88%).

Poor outcome of late acute PJI may relate to the hematogenous origin with seeding of inaccessible parts of the prosthesis like the stem which cannot be surgically debrided possibly resulting in more treatment failure.

Taken together, this review and meta-analysis found that the outcome of staphylococcal PJI after DAIR is largely determined by the type of joint and the type of causative micro-organism. Outcome for MRSA PJI seems to equal outcome for MSSA PJI. Use of rifampicin was associated with a 10% increase in success rate, but studies were hampered by confounding, publication bias and selection bias. The supporting evidence for rifampicin combination treatment is weak and possibly restricted to knee PJI, but good-quality data from randomized studies are scarce. Given this paucity of evidence, the accumulated data expose an urgent need to address the role and duration of rifampicin for staphylococcal PJI in a large randomized controlled trial.

Acknowledgments

We are indebted to Jaime Lora-Tamayo and Marjan Wouthuyzen-Bakker who kindly provided us with additional detailed information regarding rifampicin use in two cohort studies that were included in this review.

Author contributions

All authors participated in the study design, data interpretation and the writing of the manuscript and agreed to be accountable for all aspects of the work.

Funding

There was no external funding source for this study.

Potential conflicts of interest

All authors: no reported conflicts of interest.

References

1. Tande AJ, Osmon DR, Greenwood-Quaintance KE, Mabry TM, Hanssen AD, Patel R. Clinical characteristics and outcomes of prosthetic joint infection caused by small colony variant staphylococci. mBio **2014**; 5(5): e01910-14.
2. Lora-Tamayo J, Murillo O, Iribarren JA, et al. A large multicenter study of methicillin-susceptible and methicillin-resistant Staphylococcus aureus prosthetic joint infections managed with implant retention. Clin Infect Dis **2013**; 56(2): 182-94.
3. Lora-Tamayo J, Senneville E, Ribera A, et al. The Not-So-Good Prognosis of Streptococcal Periprosthetic Joint Infection Managed by Implant Retention: The Results of a Large Multicenter Study. Clin Infect Dis **2017**; 64(12): 1742-52.
4. Nguyen S, Robineau O, Titecat M, et al. Influence of daily dosage and frequency of administration of rifampicin-levofloxacin therapy on tolerance and effectiveness in 154 patients treated for prosthetic joint infections. Eur J Clin Microbiol Infect Dis **2015**; 34(8): 1675-82.
5. Vollmer NJ, Rivera CG, Stevens RW, et al. Safety and Tolerability of Fluoroquinolones in Patients with Staphylococcal Periprosthetic Joint Infections. Clin Infect Dis **2021**.
6. Osmon DR, Berbari EF, Berendt AR, et al. Diagnosis and management of prosthetic joint infection: clinical practice guidelines by the Infectious Diseases Society of America. Clin Infect Dis **2013**; 56(1): e1-e25.
7. Pijls BG, Dekkers OM, Middeldorp S, et al. AQUILA: assessment of quality in lower limb arthroplasty. An expert Delphi consensus for total knee and total hip arthroplasty. BMC Musculoskelet Disord **2011**; 12: 173.
8. Lowik CAM, Parvizi J, Jutte PC, et al. Debridement, antibiotics and implant retention is a viable treatment option for early periprosthetic joint infection presenting more than four weeks after index arthroplasty. Clin Infect Dis **2019**.
9. Wouthuyzen-Bakker M, Sebillotte M, Huotari K, et al. Lower Success Rate of Debridement and Implant Retention in Late Acute versus Early Acute Periprosthetic Joint Infection Caused by Staphylococcus spp. Results from a Matched Cohort Study. Clin Orthop Relat Res **2020**; 478(6): 1348-55.
10. Aboltins CA, Page MA, Buising KL, et al. Treatment of staphylococcal prosthetic joint infections with debridement, prosthesis retention and oral rifampicin and fusidic acid. Clin Microbiol Infect **2007**; 13(6): 586-91.
11. Lesens O, Ferry T, Forestier E, et al. Should we expand the indications for the DAIR (debridement, antibiotic therapy, and implant retention) procedure for Staphylococcus aureus prosthetic joint infections? A multicenter retrospective study. Eur J Clin Microbiol Infect Dis **2018**.
12. Joulie D, Girard J, Mares O, et al. Factors governing the healing of Staphylococcus aureus infections following hip and knee prosthesis implantation: a retrospective study of 95 patients. Orthop Traumatol Surg Res **2011**; 97(7): 685-92.
13. Dx Duffy S, Ahearn N, Darley ES, Porteous AJ, Murray JR, Howells NR. Analysis Of The KLIC-score; An Outcome Predictor Tool For Prosthetic Joint Infections Treated With Debridement, Antibiotics And Implant Retention. Journal of bone and joint infection **2018**; 3(3): 150-5.
14. Sendi P, Banderet F, Graber P, Zimmerli W. Clinical comparison between exogenous and haematogenous periprosthetic joint infections caused by Staphylococcus aureus. Clinical microbiology and infection : the official publication of the European Society of Clinical Microbiology and Infectious Diseases **2011**; 17(7): 1098-100.

15. Bouaziz A, Uckay I, Lustig S, et al. Non-compliance with IDSA guidelines for patients presenting with methicillin-susceptible Staphylococcus aureus prosthetic joint infection is a risk factor for treatment failure. Med Mal Infect **2018**; 48(3): 207-11.
16. Shohat N, Goswami K, Tan TL, Fillingham Y, Parvizi J. Increased Failure After Irrigation and Debridement for Acute Hematogenous Periprosthetic Joint Infection. J Bone Joint Surg Am **2019**; 101(8): 696-703.
17. Peel TN, Buising KL, Dowsey MM, et al. Outcome of debridement and retention in prosthetic joint infections by methicillin-resistant staphylococci, with special reference to rifampin and fusidic acid combination therapy. Antimicrob Agents Chemother **2013**; 57(1): 350-5.
18. Moojen DJ, Zwiers JH, Scholtes VA, Verheyen CC, Poolman RW. Similar success rates for single and multiple debridement surgery for acute hip arthroplasty infection. Acta Orthop **2014**; 85(4): 383-8.
19. Holmberg A, Thorhallsdottir VG, Robertsson O, Dahl A, Stefansdottir A. 75% success rate after open debridement, exchange of tibial insert, and antibiotics in knee prosthetic joint infections. Acta Orthop **2015**: 1-6.
20. Vilchez F, Martinez-Pastor JC, Garcia-Ramiro S, et al. Outcome and predictors of treatment failure in early post-surgical prosthetic joint infections due to Staphylococcus aureus treated with debridement. Clin Microbiol Infect **2011**; 17(3): 439-44.
21. Karlsen OE, Borgen P, Bragnes B, et al. Rifampin combination therapy in staphylococcal prosthetic joint infections: a randomized controlled trial. J Orthop Surg Res **2020**; 15(1): 365.
22. Aydin O, Ergen P, Ozturan B, Ozkan K, Arslan F, Vahaboglu H. Rifampin-accompanied antibiotic regimens in the treatment of prosthetic joint infections: a frequentist and Bayesian meta-analysis of current evidence. Eur J Clin Microbiol Infect Dis **2020**.
23. Morata L, Senneville E, Bernard L, et al. A Retrospective Review of the Clinical Experience of Linezolid with or Without Rifampicin in Prosthetic Joint Infections Treated with Debridement and Implant Retention. Infect Dis Ther **2014**; 3(2): 235-43.
24. Puhto AP, Puhto T, Niinimaki T, Ohtonen P, Leppilahti J, Syrjala H. Predictors of treatment outcome in prosthetic joint infections treated with prosthesis retention. Int Orthop **2015**; 39(9): 1785-91.
25. Chaussade H, Uckay I, Vuagnat A, et al. Antibiotic therapy duration for prosthetic joint infections treated by Debridement and Implant Retention (DAIR): Similar long-term remission for 6 weeks as compared to 12 weeks. International journal of infectious diseases : IJID : official publication of the International Society for Infectious Diseases **2017**; 63: 37-42.
26. Ascione T, Pagliano P, Mariconda M, et al. Factors related to outcome of early and delayed prosthetic joint infections. J Infect **2015**; 70(1): 30-6.
27. Becker A, Kreitmann L, Triffaut-Fillit C, et al. Duration of rifampin therapy is a key determinant of improved outcomes in early-onset acute prosthetic joint infection due to Staphylococcus treated with a debridement, antibiotics and implant retention (DAIR): a retrospective multicenter study in France. Journal of bone and joint infection **2020**; 5(1): 28-34.
28. Scheper H, de Boer MGJ. Comment on "Duration of rifampin therapy is a key determinant of improved outcomes in early-onset acute prosthetic joint infection due to Staphylococcus treated with a debridement, antibiotics and implant retention (DAIR): a retrospective multicenter study in France" by Becker et al. (2020). Journal of bone and joint infection **2020**; 6(1): 17-8.

29. Zimmerli W, Widmer AF, Blatter M, Frei R, Ochsner PE. Role of rifampin for treatment of orthopedic implant-related staphylococcal infections: a randomized controlled trial. Foreign-Body Infection (FBI) Study Group. JAMA **1998**; 279(19): 1537-41.
30. Scheper H, van Hooven D, van de Sande M, et al. Outcome of acute staphylococcal prosthetic joint infection treated with debridement, implant retention and antimicrobial treatment with short duration of rifampicin. The Journal of infection **2018**; 76(5): 498-500.
31. Achermann Y, Eigenmann K, Ledergerber B, et al. Factors associated with rifampin resistance in staphylococcal periprosthetic joint infections (PJI): a matched case-control study. Infection **2013**; 41(2): 431-7.
32. Hellebrekers P, Verhofstad MHJ, Leenen LPH, Varol H, van Lieshout EMM, Hietbrink F. The effect of early broad-spectrum versus delayed narrow-spectrum antibiotic therapy on the primary cure rate of acute infection after osteosynthesis. Eur J Trauma Emerg Surg **2020**; 46(6): 1341-50.
33. El Helou OC, Berbari EF, Lahr BD, et al. Efficacy and safety of rifampin containing regimen for staphylococcal prosthetic joint infections treated with debridement and retention. Eur J Clin Microbiol Infect Dis **2010**; 29(8): 961-7.

Supplementary Data

Supplemental table 1. Search strategy

Databases:	Pubmed, Embase, Cochrane Library
Search terms:	((“Debridement”[Mesh] OR “debridement”[tw] OR debrid*[tw] OR “DAIR”[tw] OR “debridement, antibiotics and implant retention”[tw] OR “debridement, antibiotics and implant retention dair”[tw] OR “debridement, antibiotics, and implant retention”[tw] OR “implant retention”[tw]) AND (“Prosthesis-Related Infections”[mesh] OR “Prosthesis Infection”[tw] OR “Prosthesis Infections”[tw] OR “Prosthetic Infection”[tw] OR “Prosthetic Infections”[tw] OR “Prosthetic Joint Infection”[tw] OR “Prosthetic Joint Infections”[tw] OR “Prosthesis-Related Infections”[tw] OR “Prosthesis-Related Infection”[tw] OR “peri prosthetic joint infection”[tw] OR “peri prosthetic joint infections”[tw] OR “periprosthetic joint infection”[tw] OR “periprosthetic joint infections”[tw] OR (“Joint Prosthesis”[Mesh] OR “Arthroplasty, Replacement”[Mesh]) AND (“Infection”[mesh] OR infect*[tw] OR “deep infection”[tw] OR “Wound Infection”[mesh] OR “Sepsis”[mesh] OR “Surgical Wound Infection”[mesh])) OR ((“Prosthesis”[tw] OR prosthe*[tw]) AND (“Joint”[tw] OR “Joints”[tw] OR “Joints”[Mesh] OR “knee”[tw] OR “shoulder”[tw] OR “elbow”[tw] OR “hip”[tw] OR “knees”[tw] OR “shoulders”[tw] OR “elbows”[tw] OR “hips”[tw]) AND (“Infection”[mesh] OR infect*[tw] OR “deep infection”[tw] OR “Wound Infection”[mesh] OR “Sepsis”[mesh] OR “Surgical Wound Infection”[mesh]))) AND (“succes rate”[tw] OR ”success rates”[tw] OR ”success”[tw] OR succes*[tw] OR “failure rate”[tw] OR ”failure rates”[tw] OR ”failure”[tw] OR fail*[tw] OR ”infection control”[tw] OR ”Treatment Outcome”[mesh] OR ”Treatment Outcome”[tw] OR ”outcome”[tw] OR ”outcomes”[tw]))

Supplemental table 2. Selected studies for systematic review

Included studies (n=64)	Year	Type of study	n	%cure total	CNS	Saureus	Knee	Hip	Cure kneePJI	Cure hipPJI	Followup
Lora-Tamayo[1]	2013	R	345	56%	NA	345	195	146	65%	66%	NA
Lora-Tamayo[2]	2013	R	44	55%	NA	44	NA	NA	NA	NA	<1yr
Shohat[3]	2019	R	113	62%	NA	113	NA	NA	NA	NA	>1yr
Duque[4]	2017	R	29	52%	NA	NA	29	0	52%	NA	>1y
Bedair[5]	2020	R	156	51%	NA	NA	92	64	44%	61%	>2y
Azzam[6]	2010	R	59	32%	NA	NA	NA	NA	NA	NA	>1y
Cobo[7]	2011	P	43	58%	NA	-/43	NA	NA	NA	NA	>1y
Fehring[8]	2013	R	63	37%	NA	NA	NA	NA	NA	NA	>2y
Theis[9]	2007	R	33	36%	NA	NA	11	22	NA	NA	NA
Senneville[10]	2011	R	41	78%	NA	NA	NA	NA	NA	NA	>3y
Tornero[11]	2015	R	176	80%	95	81	NA	NA	72%	85%	NA
Buller[12]	2012	R	182	41%	75	113	NA	NA	NA	NA	>1y
Tornero[13]	2012	R	106	76%	49	57	67	39	NA	NA	>2y
Tschudin-Sutter[14]	2016	P	81	90%	43	38	25	57	NA	NA	>2yr
Barberan[15]	2006	R	60	65%	39	21	28	32	57%	72%	>1y
Holmberg[16]	2015	R	86	74%	33	53	86	0	74%	NA	>2y
Morata[17]	2014	R	42	64%	33	9	NA	NA	NA	NA	>1yr
Wouthuyzen-Bakker[18]	2019	R	163	45%	30	141	120	44	NA	NA	NA
El Helou[19]	2010	R	87	64%	30	57	NA	NA	NA	NA	<1y
Koyonos[20]	2011	R	84	23%	28	56	NA	NA	NA	NA	>1y
Jacobs[21]	2019	R	56	79%	27	29	NA	NA	NA	NA	>1y
Wouthuyzen-Bakker[22]	2020	R	180	56%	26	154	91	62	NA	NA	>1y

Supplemental table 2. Continued

Included studies (n=64)	Year	Type of study	n	%cure total	CNS	Saureus	Knee	Hip	Cure kneePJI	Cure hipPJI	Followup
Byren[23]	2009	R	73	75%	26	47	NA	NA	NA	NA	NA
Chaussade[24]	2017	R	37	59%	25	12	NA	NA	n	NA	>1y
Fink[25]	2017	R	49	72%	25	24	NA	NA	NA	NA	>2y
Marculescu[26]	2006	R	55	33%	23	32	NA	NA	NA	NA	>1yr
Grammatopoulos[27]	2017	R	61	87%	22	39	0	61	NA	87%	>1y
Bryan[28]	2017	R	58	84%	21	37	0	58	NA	84%	>2y
Peel[29]	2013	R	43	79%	19	24	15	28	93%	71%	>1yr
Parvizi[30]	2009	R	35	37%	18	17	11	24	45%	33%	>1yr
Koh[31]	2015	R	27	70%	16	11	27	0	70%	NA	>2y
Becker[32]	2020	R	79	68%	16	65	21	59	71%	68%	>2y
Zmistowski[33]	2016	R	81	48%	16	65	NA	NA	NA	NA	>1yr
Weenders[34]	2016	R	21	82%	15	7	0	21	NA	82%	>2y
Lora-Tamayo[35]	2016	RCT	63	65%	15	48	34	29	46%	58%	>1yr
Triantafyllopoulos[36]	2015	R	55	45%	14	41	55	0	45%	NA	>1yr
Soriano[37]	2006	R	25	76%	14	11	11	14	45%	100%	>1yr
Kuiper[38]	2013	R	63	60%	13	50	NA	NA	NA	NA	>1y
Dx Duffy[39]	2018	R	33	76%	12	21	33	0	76%	NA	>2y
Lizaur-Utrilla[40]	2015	R	27	26%	11	16	27	0	26%	NA	>1yr
Sendi[41]	2017	P	21	90%	11	10	0	21	NA	90%	>2y
Moojen[42]	2014	R	50	80%	11	39	0	50	NA	80%	>1yr
Swenson[43]	2018	R	40	58%	11	29	NA	NA	NA	NA	>6m
Gardner[44]	2011	R	27	44%	10	17	27	0	44%	NA	NA
Karlsen[45]	2020	RCT	48	73%	10	38	9	39	73%	67%	>2y
Ottesen[46]	2019	R	37	89%	8	29	37	0	88%	NA	>1yr
Zhang[47]	2017	R	23	26%	5	18	23	0	26%	NA	>1yr
Flierl[48]	2017	R	20	50%	4	16	NA	NA	NA	NA	>1y
Waagsbo[49]	2009	R	26	77%	3	23	0	26	NA	NA	>2y
Scheper[50]	2018	R	41	63%	11	30	19	22	44%	83%	>6m
Kuo[51]	2019	R	26	50%	2	24	NA	NA	NA	NA	>1y
Aboltins[52]	2007	R	20	90%	1	19	7	13	86%	92%	>1y
Wilson[53]	1990	R	22	41%	0	22	22	0	41%	NA	>1yr
Bene (2x:H+K)[54, 55]	2018	R	21	62%	0	21	15	6	53%	83%	>2y
Brandt[56]	1997	R	33	36%	0	33	26	7	38%	29%	>1y
Vilchez[57]	2011	R	53	75%	0	53	35	18	69%	89%	>1yr
Wouthuyzen-Bakker[58]	2018	R	58	88%	0	58	34	24	NA	NA	>3y
Betz[59]	2015	R	29	76%	0	29	0	29	NA	76%	>3y
Bouaziz[60]	2018	R	89	58%	0	89	35	54	51%	63%	>1y
Lesens[61]	2018	R	137	76%	0	137	57	77	NA	NA	>1y
Letouvet[62]	2016	R	24	50%	0	24	NA	NA	NA	NA	>1y
Löwik[63]	2019	R	339	54%	0	339	NA	NA	NA	NA	>1yr
Hirsiger[64]	2019	R	29	90%	0	29	NA	NA	NA	NA	>3y
Joulie[65]	2011	R	53	57%	0	53	NA	NA	NA	NA	>2y

The largest included study presented outcome for both *S. aureus* and coagulase-negative staphylococci (CNS) but due to many polymicrobial infections, we decided to leave out CNS from this study in the analysis to prevent a large group of duplicate outcomes[63].

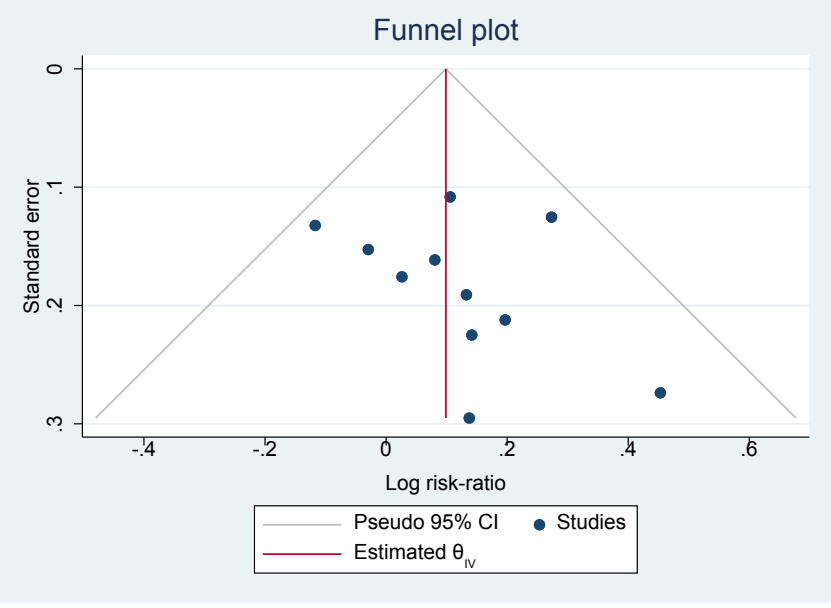
Supplemental table 3. Newcastle-Ottawa Quality assessment scale of included studies in meta-analysis

First author	Year of publication	Selection	Comparability	Outcome	Quality
Holmberg[16]	2015	****	-	***	Poor
Chaussade[24]	2017	****	**	***	Good
El Helou[19]	2010	***	-	***	Poor
Lora-Tamayo[2]	2013	****	**	**	Good
Senneville[10]	2011	****	-	**	Poor
Tornero[13]	2012	****	-	***	Poor
Vilchez[57]	2011	****	**	***	Good
Wouthuyzen-Bakker[18]	2019	****	**	***	Good
Becker[32]	2020	****	**	***	Good
Karlsen[45]	2020	****	**	***	Good
Bryan[28]	2017	****	*	***	Good

Scoring for comparability: one star was given if a regression analysis was performed including rifampicin AND type of joint or type as dependent variables, two stars were given if also other variables were included (proportion of S aureus PJI, age, comorbidity index)

Quality was registered according to Newcastle-Otawa Quality criteria:

- **Good quality**: 3 or 4 stars in selection domain AND 1 or 2 stars in comparability domain AND 2 or 3 stars in outcome/exposure domain.
- **Fair quality**: 2 stars in selection domain AND 1 or 2 stars in comparability domain AND 2 or 3 stars in outcome/exposure domain.
- **Poor quality**: 0 or 1 star in selection domain OR 0 stars in comparability domain OR 0 or 1 stars in outcome/exposure domain.



Supplemental figure 1. Funnel plot of 11 included studies in meta-analysis

Supplemental table 4. Outcome of staphylococcal PJI after DAIR in 30 studies that reported individual patient data regarding the use of rifampicin or not.

Type of joint	Rifampicin N cured/N total (%)	No rifampicin N cured/N total (%)	RR for success with rifampicin (95% CI)
Hip PJI			
Bryan	26/29 (90%)	25/31(81%)	1.11(0.90-1.37)
Moojen	37/47 (79%)	3/3 (100%)	
Waagsbo	20/26 (77%)		
Sendi	19/21 (90%)		
Total hip PJI	**102/123 (83%)**	**28/34 (82%)**	**1.01 (0.85-2.00)**
Knee PJI			
Holmberg	56/69 (81%)	8/12 (67%)	1.22 (0.80-1.85)
Wilson		9/22 (41%)	
Total knee PJI	**56/69 (81%)**	**17/34 (50%)**	**1.62 (1.14-2.31)**
Hip and Knee PJI			
Peel	31/40 (78%)	3/3 (100%)	
Wouthuyzen-Bakker 2019	68/134 (50%)	10/31 (32%)	1.57 (0.92-2.69)
Tornero	70/93 (75%)	11/13 (85%)	0.88 (0.69-1.15)
Vilchez	37/43 (86%)	4/10 (40%)	1.15 (0.64-2.05)
Senneville	25/31(81%)	7/10(70%)	1.15 (0.74-1.79)
El Helou	21/31(68%)	35/56(63%)	1.08 (0.79-1.49)
Lora-Tamayo 2013	146/216 (68%)	36/70 (51%)	1.31 (1.03-1.68)
Karlsen	17/23 (74%)	18/25 (72%)	1.03 (0.73-1.45)
Becker	41/58 (71%)	13/21 (62)%	1.14 (0.79-1.66)
Chaussade	41/60 (68%)	19/27(70%)	0.97(0.72-1.31)
Marculescu		18/55(33%)	
Brandt		12/33(36%)	
Lora-Tamayo 2016	41/63 (65%)		
Wouthuyzen-Bakker 2018	51/58 (88%)		
Barberan	39/60 (65%)		
Tschudin-Sutter	73/81(90%)		
Jacobs	44/56(79%)		
Cobo	35/57(61%)		
Fink	35/49(71%)		
Parvizi	13/35 (25%)		
Aboltins	18/20 (90%)		
Soriano	19/25 (76%		
Scheper	26/41 (63%)		
Letouvet	12/24(50%)		
Total hip+knee PJI	**903/1298 (70%)**	**186/354 (53%)**	**1.32 (1.19-1.47)**
Total	**1061/1490 (71%)**	**231/422 (55%)**	**1.30 (1.86-1.43)**

DAIR: debridement, antibiotics, implant retention
PJI: prosthetic joint infection

References

1. Lora-Tamayo J, Murillo O, Iribarren JA, et al. A large multicenter study of methicillin-susceptible and methicillin-resistant Staphylococcus aureus prosthetic joint infections managed with implant retention. Clin Infect Dis **2013**; 56(2): 182-94.
2. Lora-Tamayo J, Euba G, Ribera A, et al. Infected hip hemiarthroplasties and total hip arthroplasties: Differential findings and prognosis. The Journal of infection **2013**; 67(6): 536-44.
3. Shohat N, Goswami K, Tan TL, Fillingham Y, Parvizi J. Increased Failure After Irrigation and Debridement for Acute Hematogenous Periprosthetic Joint Infection. J Bone Joint Surg Am **2019**; 101(8): 696-703.
4. Duque AF, Post ZD, Lutz RW, Orozco FR, Pulido SH, Ong AC. Is There Still a Role for Irrigation and Debridement With Liner Exchange in Acute Periprosthetic Total Knee Infection? J Arthroplasty **2017**; 32(4): 1280-4.
5. Bedair HS, Katakam A, Bedeir YH, Yeroushalmi D, Schwarzkopf R. A decision analysis of treatment strategies for acute periprosthetic joint infection: Early irrigation and debridement versus delayed treatment based on organism. J Orthop **2020**; 22: 246-50.
6. Azzam KA, Seeley M, Ghanem E, Austin MS, Purtill JJ, Parvizi J. Irrigation and debridement in the management of prosthetic joint infection: traditional indications revisited. J Arthroplasty **2010**; 25(7): 1022-7.
7. Cobo J, Miguel LG, Euba G, et al. Early prosthetic joint infection: outcomes with debridement and implant retention followed by antibiotic therapy. Clinical microbiology and infection : the official publication of the European Society of Clinical Microbiology and Infectious Diseases **2011**; 17(11): 1632-7.
8. Fehring TK, Odum SM, Berend KR, et al. Failure of irrigation and debridement for early postoperative periprosthetic infection. Clin Orthop Relat Res **2013**; 471(1): 250-7.
9. Theis JC, Gambhir S, White J. Factors affecting implant retention in infected joint replacements. ANZ journal of surgery **2007**; 77(10): 877-9.
10. Senneville E, Joulie D, Legout L, et al. Outcome and predictors of treatment failure in total hip/knee prosthetic joint infections due to Staphylococcus aureus. Clin Infect Dis **2011**; 53(4): 334-40.
11. Tornero E, Morata L, Martinez-Pastor JC, et al. KLIC-score for predicting early failure in prosthetic joint infections treated with debridement, implant retention and antibiotics. Clinical microbiology and infection : the official publication of the European Society of Clinical Microbiology and Infectious Diseases **2015**; 21(8): 786 e9- e17.
12. Buller LT, Sabry FY, Easton RW, Klika AK, Barsoum WK. The preoperative prediction of success following irrigation and debridement with polyethylene exchange for hip and knee prosthetic joint infections. J Arthroplasty **2012**; 27(6): 857-64 e1-4.
13. Tornero E, Garcia-Oltra E, Garcia-Ramiro S, et al. Prosthetic joint infections due to Staphylococcus aureus and coagulase-negative staphylococci. Int J Artif Organs **2012**; 35(10): 884-92.
14. Tschudin-Sutter S, Frei R, Dangel M, et al. Validation of a treatment algorithm for orthopaedic implant-related infections with device-retention-results from a prospective observational cohort study. Clin Microbiol Infec **2016**; 22(5).

15. Barberan J, Aguilar L, Carroquino G, et al. Conservative treatment of staphylococcal prosthetic joint infections in elderly patients. Am J Med **2006**; 119(11): 993-10.
16. Holmberg A, Thorhallsdottir VG, Robertsson O, Dahl A, Stefansdottir A. 75% success rate after open debridement, exchange of tibial insert, and antibiotics in knee prosthetic joint infections. Acta Orthop **2015**: 1-6.
17. Morata L, Senneville E, Bernard L, et al. A Retrospective Review of the Clinical Experience of Linezolid with or Without Rifampicin in Prosthetic Joint Infections Treated with Debridement and Implant Retention. Infect Dis Ther **2014**; 3(2): 235-43.
18. Wouthuyzen-Bakker M, Sebillotte M, Lomas J, et al. Clinical outcome and risk factors for failure in late acute prosthetic joint infections treated with debridement and implant retention. The Journal of infection **2019**; 78(1): 40-7.
19. El Helou OC, Berbari EF, Lahr BD, et al. Efficacy and safety of rifampin containing regimen for staphylococcal prosthetic joint infections treated with debridement and retention. Eur J Clin Microbiol Infect Dis **2010**; 29(8): 961-7.
20. Koyonos L, Zmistowski B, Della Valle CJ, Parvizi J. Infection control rate of irrigation and debridement for periprosthetic joint infection. Clin Orthop Relat Res **2011**; 469(11): 3043-8.
21. Jacobs AME, Valkering LJJ, Benard M, Meis JF, Goosen JHM. Evaluation One Year after DAIR Treatment in 91 Suspected Early Prosthetic Joint Infections in Primary Knee and Hip Arthroplasty. Journal of bone and joint infection **2019**; 4(5): 238-44.
22. Wouthuyzen-Bakker M, Sebillotte M, Huotari K, et al. Lower Success Rate of Debridement and Implant Retention in Late Acute versus Early Acute Periprosthetic Joint Infection Caused by Staphylococcus spp. Results from a Matched Cohort Study. Clin Orthop Relat Res **2020**; 478(6): 1348-55.
23. Byren I, Bejon P, Atkins BL, et al. One hundred and twelve infected arthroplasties treated with 'DAIR' (debridement, antibiotics and implant retention): antibiotic duration and outcome. J Antimicrob Chemother **2009**; 63(6): 1264-71.
24. Chaussade H, Uckay I, Vuagnat A, et al. Antibiotic therapy duration for prosthetic joint infections treated by Debridement and Implant Retention (DAIR): Similar long-term remission for 6 weeks as compared to 12 weeks. International journal of infectious diseases : IJID : official publication of the International Society for Infectious Diseases **2017**; 63: 37-42.
25. Fink B, Schuster P, Schwenninger C, Frommelt L, Oremek D. A Standardized Regimen for the Treatment of Acute Postoperative Infections and Acute Hematogenous Infections Associated With Hip and Knee Arthroplasties. J Arthroplasty **2017**; 32(4): 1255-61.
26. Marculescu CE, Berbari EF, Hanssen AD, et al. Outcome of prosthetic joint infections treated with debridement and retention of components. Clin Infect Dis **2006**; 42(4): 471-8.
27. Grammatopoulos G, Kendrick B, McNally M, et al. Outcome Following Debridement, Antibiotics, and Implant Retention in Hip Periprosthetic Joint Infection-An 18-Year Experience. J Arthroplasty **2017**; 32(7): 2248-55.

28. Bryan AJ, Abdel MP, Sanders TL, Fitzgerald SF, Hanssen AD, Berry DJ. Irrigation and Debridement with Component Retention for Acute Infection After Hip Arthroplasty: Improved Results with Contemporary Management. J Bone Joint Surg Am **2017**; 99(23): 2011-8.
29. Peel TN, Buising KL, Dowsey MM, et al. Outcome of debridement and retention in prosthetic joint infections by methicillin-resistant staphylococci, with special reference to rifampin and fusidic acid combination therapy. Antimicrob Agents Chemother **2013**; 57(1): 350-5.
30. Parvizi J, Azzam K, Ghanem E, Austin MS, Rothman RH. Periprosthetic infection due to resistant staphylococci: serious problems on the horizon. Clin Orthop Relat Res **2009**; 467(7): 1732-9.
31. Koh IJ, Han SB, In Y, et al. Open debridement and prosthesis retention is a viable treatment option for acute periprosthetic joint infection after total knee arthroplasty. Archives of orthopaedic and trauma surgery **2015**; 135(6): 847-55.
32. Becker A, Kreitmann L, Triffaut-Fillit C, et al. Duration of rifampin therapy is a key determinant of improved outcomes in early-onset acute prosthetic joint infection due to Staphylococcus treated with a debridement, antibiotics and implant retention (DAIR): a retrospective multicenter study in France. Journal of bone and joint infection **2020**; 5(1): 28-34.
33. Zmistowski BM, Manrique J, Patel R, Chen AF. Recurrent Periprosthetic Joint Infection After Irrigation and Debridement With Component Retention Is Most Often Due to Identical Organisms. J Arthroplasty **2016**; 31(9 Suppl): 148-51.
34. Weenders SGM, Nijhof MW, Schimmel JJP, Goosen JHM. Debridement, antibiotics and implant retention in early periprosthetic joint infection after primary total hip arthroplasty : 88 percent survival after two years follow-up. Acta orthopaedica Belgica **2016**; 82(3): 530-8.
35. Lora-Tamayo J, Euba G, Cobo J, et al. Short- versus long-duration levofloxacin plus rifampicin for acute staphylococcal prosthetic joint infection managed with implant retention: a randomised clinical trial. International journal of antimicrobial agents **2016**; 48(3): 310-6.
36. Triantafyllopoulos GK, Poultsides LA, Zhang W, Sculco PK, Ma Y, Sculco TP. Periprosthetic knee infections treated with irrigation and debridement: outcomes and preoperative predictive factors. J Arthroplasty **2015**; 30(4): 649-57.
37. Soriano A, Garcia S, Bori G, et al. Treatment of acute post-surgical infection of joint arthroplasty. Clin Microbiol Infect **2006**; 12(9): 930-3.
38. Kuiper JW, Vos SJ, Saouti R, et al. Prosthetic joint-associated infections treated with DAIR (debridement, antibiotics, irrigation, and retention): analysis of risk factors and local antibiotic carriers in 91 patients. Acta Orthop **2013**; 84(4): 380-6.
39. Dx Duffy S, Ahearn N, Darley ES, Porteous AJ, Murray JR, Howells NR. Analysis Of The KLIC-score; An Outcome Predictor Tool For Prosthetic Joint Infections Treated With Debridement, Antibiotics And Implant Retention. Journal of bone and joint infection **2018**; 3(3): 150-5.
40. Lizaur-Utrilla A, Gonzalez-Parreno S, Gil-Guillen V, Lopez-Prats FA. Debridement with prosthesis retention and antibiotherapy vs. two-stage revision for periprosthetic knee infection within 3 months after arthroplasty: a case-control study. Clinical microbiology and infection : the official publication of the European Society of Clinical Microbiology and Infectious Diseases **2015**; 21(9): 851 e11-7.

41. Sendi P, Lotscher PO, Kessler B, Graber P, Zimmerli W, Clauss M. Debridement and implant retention in the management of hip periprosthetic joint infection: outcomes following guided and rapid treatment at a single centre. Bone Joint J **2017**; 99-B(3): 330-6.
42. Moojen DJ, Zwiers JH, Scholtes VA, Verheyen CC, Poolman RW. Similar success rates for single and multiple debridement surgery for acute hip arthroplasty infection. Acta Orthop **2014**; 85(4): 383-8.
43. Swenson RD, Butterfield JA, Irwin TJ, Zurlo JJ, Davis CM, 3rd. Preoperative Anemia Is Associated With Failure of Open Debridement Polyethylene Exchange in Acute and Acute Hematogenous Prosthetic Joint Infection. J Arthroplasty **2018**; 33(6): 1855-60.
44. Gardner J, Gioe TJ, Tatman P. Can this prosthesis be saved?: implant salvage attempts in infected primary TKA. Clin Orthop Relat Res **2011**; 469(4): 970-6.
45. Karlsen OE, Borgen P, Bragnes B, et al. Rifampin combination therapy in staphylococcal prosthetic joint infections: a randomized controlled trial. J Orthop Surg Res **2020**; 15(1): 365.
46. Ottesen CS, Troelsen A, Sandholdt H, Jacobsen S, Husted H, Gromov K. Acceptable Success Rate in Patients With Periprosthetic Knee Joint Infection Treated With Debridement, Antibiotics, and Implant Retention. J Arthroplasty **2019**; 34(2): 365-8.
47. Zhang C, Yan CH, Chan PK, Ng FY, Chiu KY. Polyethylene Insert Exchange Is Crucial in Debridement for Acute Periprosthetic Infections following Total Knee Arthroplasty. J Knee Surg **2017**; 30(1): 36-41.
48. Flierl MA, Culp BM, Okroj KT, Springer BD, Levine BR, Della Valle CJ. Poor Outcomes of Irrigation and Debridement in Acute Periprosthetic Joint Infection With Antibiotic-Impregnated Calcium Sulfate Beads. J Arthroplasty **2017**; 32(8): 2505-7.
49. Waagsbo B, Sundoy A, Martinsen TM, Nymo LS. Treatment results with debridement and retention of infected hip prostheses. Scand J Infect Dis **2009**; 41(8): 563-8.
50. Scheper H, van Hooven D, Visser L, et al. Outcome of acute staphylococcal prosthetic joint infection treated with debridement, implant retention and antimicrobial treatment with short duration of rifampicin. The Journal of infection **2018**.
51. Kuo FC, Goswami K, Klement MR, Shohat N, Parvizi J. Positive Blood Cultures Decrease the Treatment Success in Acute Hematogenous Periprosthetic Joint Infection Treated With Debridement, Antibiotics, and Implant Retention. J Arthroplasty **2019**.
52. Aboltins CA, Page MA, Buising KL, et al. Treatment of staphylococcal prosthetic joint infections with debridement, prosthesis retention and oral rifampicin and fusidic acid. Clin Microbiol Infect **2007**; 13(6): 586-91.
53. Wilson MG, Kelley K, Thornhill TS. Infection as a complication of total knee-replacement arthroplasty. Risk factors and treatment in sixty-seven cases. J Bone Joint Surg Am **1990**; 72(6): 878-83.
54. Bene N, Li X, Nandi S. Increased antibiotic duration improves reoperation free survival after total hip arthroplasty irrigation and debridement. J Orthop **2018**; 15(2): 707-10.
55. Bene N, Li X, Nandi S. Factors affecting failure of irrigation and debridement with liner exchange in total knee arthroplasty infection. Knee **2018**; 25(5): 932-8.
56. Brandt CM, Sistrunk WW, Duffy MC, et al. Staphylococcus aureus prosthetic joint infection treated with debridement and prosthesis retention. Clin Infect Dis **1997**; 24(5): 914-9.

57. Vilchez F, Martinez-Pastor JC, Garcia-Ramiro S, et al. Outcome and predictors of treatment failure in early post-surgical prosthetic joint infections due to Staphylococcus aureus treated with debridement. Clin Microbiol Infect **2011**; 17(3): 439-44.
58. Wouthuyzen-Bakker M, Tornero E, Morata L, et al. Moxifloxacin plus rifampin as an alternative for levofloxacin plus rifampin in the treatment of a prosthetic joint infection with Staphylococcus aureus. International journal of antimicrobial agents **2018**; 51(1): 38-42.
59. Betz M, Abrassart S, Vaudaux P, et al. Increased risk of joint failure in hip prostheses infected with Staphylococcus aureus treated with debridement, antibiotics and implant retention compared to Streptococcus. Int Orthop **2015**; 39(3): 397-401.
60. Bouaziz A, Uckay I, Lustig S, et al. Non-compliance with IDSA guidelines for patients presenting with methicillin-susceptible Staphylococcus aureus prosthetic joint infection is a risk factor for treatment failure. Med Mal Infect **2018**; 48(3): 207-11.
61. Lesens O, Ferry T, Forestier E, et al. Should we expand the indications for the DAIR (debridement, antibiotic therapy, and implant retention) procedure for Staphylococcus aureus prosthetic joint infections? A multicenter retrospective study. Eur J Clin Microbiol Infect Dis **2018**.
62. Letouvet B, Arvieux C, Leroy H, et al. Predictors of failure for prosthetic joint infections treated with debridement. Med Mal Infect **2016**; 46(1): 39-43.
63. Lowik CAM, Parvizi J, Jutte PC, et al. Debridement, antibiotics and implant retention is a viable treatment option for early periprosthetic joint infection presenting more than four weeks after index arthroplasty. Clin Infect Dis **2019**.
64. Hirsiger S, Betz M, Stafylakis D, Gotschi T, Lew D, Uckay I. The Benefice of Mobile Parts' Exchange in the Management of Infected Total Joint Arthroplasties with Prosthesis Retention (DAIR Procedure). J Clin Med **2019**; 8(2).
65. Joulie D, Girard J, Mares O, et al. Factors governing the healing of Staphylococcus aureus infections following hip and knee prosthesis implantation: a retrospective study of 95 patients. Orthop Traumatol Surg Res **2011**; 97(7): 685-92.

CHAPTER 4a

Rifampin for staphylococcal PJI, do we still need a randomized controlled trial?

Henk Scheper, Mark G.J. de Boer

Clinical Infectious Diseases 2022; 74: (7): 1316–1318

With great interest we read the observational study by Beldman *et al*. in which the additional value of rifampin for patients with staphylococcal prosthetic joint infection was evaluated[1]. Their data show a favorable effect of rifampin after adjustments. However, the data presented evoke the thought that the results remain flawed by confounding by indication and immortal time bias.

In general, four centers using rifampin were compared with only one center not using rifampin. Centers can be outliers with regard to PJI treatment results. Over the years, success rates after DAIR showed large variety in different cohorts, ranging between 30% and 90% (Figure 1) [2]. Taking a single center as a reference may hence distort the outcome in a way that cannot be corrected for. Furthermore, as surgical strategies certainly improved over the past 20 years, the distribution of the data over time should be taken into account.

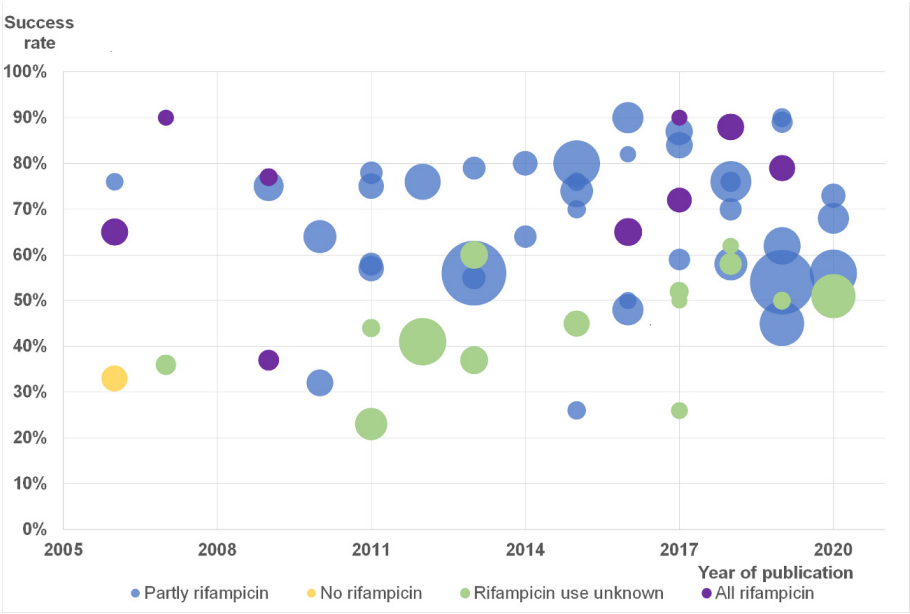


Figure 1. Success rates over the years for staphylococcal PJI treated with debridement, antibiotics, and retention of the implant (DAIR) and related to use of rifampicin (review of 64 studies) [2].

After excluding all patients who failed before switching to oral therapy, only the failure rate in the non-rifampin group dropped, from 54.2% to 45.4%. This indicates that baseline characteristics must have been substantially different (rifampin cannot explain this as it had not been started yet in both groups). It also shows the presence of immortal time bias. Hence, it would be interesting to know the outcome of a multivariate time-to-event cox regression analysis, starting on the moment of antibiotic switch.

Confounding by indication was meant to be reduced by excluding patients in 'rifampin-centers' who were not treated with rifampin, However, confounding is more likely to be induced here as there is always a reason why patients in rifampin centers are not treated with rifampin (e.g., because of early failure, because of continuing intravenous antibiotics, et cetera). In the non-rifampin center, these patients are included and may be responsible for a worse outcome.

Of note, the proportion of knee PJI in the rifampin group was lower than in the non-rifampin group (40% vs. 46%, p 0.13) which may also affect outcome.

Lastly, early start of rifampin (within 5 days after DAIR) was associated with an increased failure rate which led to the conclusion that early start should be discouraged. However, the presented data show that these early starters also had much more *S. aureus* infections (74% vs 51%), less exchange of mobile parts and later onset of DAIR after PJI diagnosis, all of which known to be associated with failure. A multivariate Cox regression analysis of early versus later start of rifampin would be insightful. The difference in failure rate may disappear after correction for the above mentioned risk factors. In that case, early start of rifampin is more an epiphenomenon rather than a risk factor for failure.

Although the association between using rifampin and success is statistically demonstrated in these pooled cohorts, confounding and immortal time bias are likely to be present. Even with multivariate analysis, proving causality is difficult, which is why a randomized controlled trial is the only way forward to solve this difficult but highly relevant clinical question.

Potential conflicts of interest.
HS: No conflicts of interest. MdB: no conflicts of interest.

References

1. Beldman M, Lowik C, Soriano A, et al. If, when, and how to use rifampin in acute staphylococcal periprosthetic joint infections, a multicentre observational study. *Clin Infect Dis* 2021 doi: 10.1093/cid/ciab426 [published Online First: 2021/05/11]

2. Scheper H, Gerritsen LM, Pijls BG et al. Outcome of Debridement, Antibiotics, and Implant Retention for Staphylococcal Hip and Knee Prosthetic Joint Infections, Focused on Rifampicin Use: A Systematic Review and Meta-Analysis. Open Forum Infect Dis. 2021 Jul 1;8(7) doi: 10.1093/ofid/ofab298.

CHAPTER 4b

Reported association between duration of rifampicin and improved outcomes in acute staphylococcal prosthetic joint infection: analysis hampered by methodological errors

Henk Scheper, Mark G.J. de Boer

J Bone Jt Infect 2020 Jul 22;6(1):17-18

Dear editor,

The adjunctive role of rifampicin for staphylococcal prosthetic joint infection is an important and ongoing discussion. We compliment our colleagues with studying this important question in a multicenter collaboration(1). The authors conclude that prolonged duration of rifampicin therapy is a key determinant for improved outcomes in acute staphylococcal prosthetic joint infection treated with DAIR. However, this conclusion seems to be flawed due to survival bias, exclusion bias and probably confounding by indication.

Survival bias is correctly mentioned by the authors. Rifampicin is often started two weeks after debridement when wounds are dry and antimicrobial sensitivity is known. All patients with early failures until start of rifampicin do not 'survive' this period and will be assigned to the non-rifampicin group, leading to a skewed selection of failures in the non-rifampicin group. Correction for this bias is challenging and could be solved through optimal use of randomization methods. There also other methods, or designs.

Confounding by indication is inevitable in retrospective studies that aim to study treatment effects. For unclear reasons, 24% of patients did not receive rifampicin, possibly because in this group drug-drug interactions or other comorbidities that may be independent risk factors for failure were present. Though authors studied other factors associated with DAIR failure (smoking, diabetes mellitus, ASA score, rifampin combination therapy with a fluoroquinolone), residual confounding remains due to these factors for which correction is difficult (e.g. by propensity score methods under the condition that the correct variables were obtained)".

The most important limitation of this study is that the authors decided to exclude DAIR failures occurring while the patient was still under rifampicin. Bias was not prevented but intentionally introduced with this measure, as only failures in the rifampicin group can be excluded. This resulted in the observation of an even more skewed positive response in the group of patients receiving rifampicin.

Taken together, the results of this study should lead to a more cautious conclusion. Duration of rifampicin is associated with better outcome but this effect may be solely explained by introduction of bias by removing patients that failed on rifampicin treatment, confounding by indication and survival bias. Figure 1 shows how bias can potentially lead to erroneous conclusions in this type of observational cohort studies. It is important to address these issues and to correct for them as much as possible upfront.

We completely agree with the authors that high-quality studies are warranted to elucidate the optimal duration of rifampicin as part of the antimicrobial therapy in patients with a staphylococcal PJI. A randomized controlled trial can answer the important question about the optimal duration of adjunctive use of rifampicin for staphylococcal PJI.

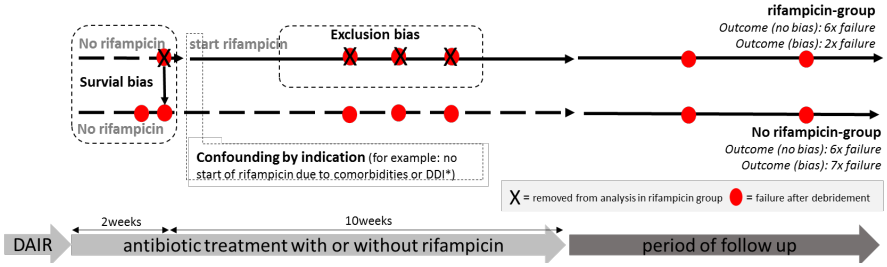


Figure 1. Hypothetical example of a PJI study with a flawed outcome induced by methodological errors.

Example of a PJI cohort, retrospectively stratified by use of rifampin. Survival bias occurs because only patients that 'survive' the first weeks until start with rifampin are analyzed in rifampin-group. All failures before start of rifampin will be analyzed in the non-rifampin group. Confounding by indication occurs when patients with certain risk factors for failure (e.g. comorbidities, drug-drug interactions, severely ill) are not selected for treatment with rifampin. Exclusion bias occurs if patients are excluded while they still use rifampin, as only failures within the rifampin group can be excluded. In this hypothetical example assuming comparable treatment strategies, both groups would have an identical failure rate without bias (both 6 failures), but three times as much failure in the non-rifampin group after introduction of bias.

*DDI: drug-drug interaction, f/u: follow up. DAIR: debridement, antibiotics, implant retention.

References

1. Becker A, Kreitmann L, Triffaut-Fillit C, Valour F, Mabrut E, Forestier E, et al. Duration of rifampin therapy is a key determinant of improved outcomes in early-onset acute prosthetic joint infection due to Staphylococcus treated with a debridement, antibiotics and implant retention (DAIR): a retrospective multicenter study in France. Journal of bone and joint infection. 2020;5(1):28-34.

CHAPTER 5

Infected tumor prostheses of the lower extremities: causative micro-organisms, effectiveness of DAIR and risk factors for treatment failure

Philip T.J. Sanders, Henk Scheper, Robert J.P. van der Wal, Michiel A.J. van de Sande, Mark G.J. de Boer, Sander D.S. Dijkstra, Michael P.A. Bus

Journal of Bone & Joint Surgery, *under review*

Background

Infection of tumor endoprostheses after reconstruction of the lower extremities is a common complication and treatment of these infections is challenging and often requires multiple surgical interventions or even implant removal. Because there is limited evidence to support treatment strategies and knowledge of epidemiology of causative micro-organisms, we analyzed the effectiveness of Debridement, Antibiotics and Implant Retention (DAIR), risk factors for failure of DAIR and causative micro-organisms in patients with an infected tumor endoprosthesis of the lower extremity.

Methods

A retrospective cohort study was conducted. In a tertiary referral center for orthopedic oncology, all patients treated for prosthetic joint infection (PJI) between 2000 and 2018 with an infection of a tumor endoprosthesis of the lower extremities were included. Treatment outcomes and risk factors for failure were analyzed in patients primarily treated with DAIR. Causative micro-organisms were recorded. The minimum follow-up period was two years.

Results

Of 337 patients who underwent endoprosthetic reconstruction of the lower extremities, 67 patients (20%) developed an infection of a tumor endoprosthesis. Of them, 55 were primarily treated with DAIR. The cure rate of DAIR was 65% (36/55). A median of 2 debridements per patient was needed. Chemotherapy (OR=3.1,95%CI=1.0-9.3) and erythrocyte sedimentation rate >50 at diagnosis (OR=4.5,95%CI=1.3-15.4) were associated with treatment failure. Eighteen (27%) patients had a polymicrobial infection.

Conclusions

Although sequential procedures are often needed, the DAIR-procedure has acceptable clinical outcome and should be considered dependent on expected survival and risk factors for treatment failure noted in this study.

Background

Modular endoprosthetic reconstruction is the preferred reconstructive technique after tumor resection of the lower extremities in most orthopedic oncology centers. A prosthetic joint infection (PJI) remains one of the major challenges, with reported incidences of up to 15% (table 1). These infections can be devastating, as they regularly necessitate multiple surgical debridements, removal of implants, or, rarely, amputation[1,2]. Treatment of infection often results in delayed start of chemotherapy and possibly deterioration of oncologic outcomes. Patients undergoing tumor resection and subsequent reconstruction surgery may have an increased risk of PJI due to disseminated malignancy, the use of neoadjuvant chemotherapy and radiation therapy[3]. Tumor resection and reconstruction is usually lengthy, results in large wound beds with extended soft tissue removal, and possibilities for adequate soft tissue coverage are often limited requiring vascularized muscle flaps. These factors may contribute to the marked differences of infection risk when compared to conventional arthroplasty (9-15% vs <1%) (table 2)[4,5].

Surgical treatment of an infected tumor endoprosthesis consists of debridement, antibiotics and implant retention (DAIR) or a one- or two-staged exchange of the implant. For PJI after conventional arthroplasty, the indications for the type of surgical strategy are well defined and clinical outcomes of these strategies are reported on extensively [6]. However, there is a lack of data on clinical outcomes of surgical strategies for infected tumor endoprostheses that can guide in the decision to perform either DAIR, one-stage, or two-stage revision procedures. Therefore, we analyzed (1) causative micro-organisms, (2) clinical outcome of surgical treatment strategies and (3) risk factors for treatment failure in a cohort of patients with an infected tumor endoprosthesis of the lower extremity. In addition, we reviewed the literature regarding surgical management of infected tumor endoprostheses.

Table 1. Baseline characteristics of 67 patients with tumor endoprosthesis PJI.

	n	%
Gender (male)	43	64
Localization		
Proximal femur	21	31
Distal femur	32	48
Proximal tibia	10	15
Total femur	2	3
Intercalary femur	2	3
Diagnosed bone tumor		
Osteosarcoma	24	36
Chondrosarcoma	14	21
Ewing Sarcoma	3	5
Soft tissue sarcoma	6	9
Benign tumors	8	12
Metastasis	12	18
ASA classification[$]		
ASA 1	8	12
ASA 2	45	67
ASA 3	13	19
ASA 4	1	2
Chemotherapy (adjuvant)	33	49
Radiotherapy (adjuvant)	9	13
Silver coating	20	30
Cemented fixation	24	36
Prophylactic antibiotic mats	10	15
Infection after revision procedure	33	49
Implant loosening	4	6
Fistula	6	9
Acute PJI (<6w)	29	43
Early chronic PJI (6-12w)	13	19
Chronic PJI (>12w)	25	37
Late acute (hematogenous) PJI	0	-

[$]American society of anesthesiologists classification system

Table 2. Causative micro-organisms of 67 patient's tumor endoprosthesis PJI.

Causative micro-organisms	Monomicrobial (n=37, 55%)	Polymicrobial (n=19, 30%)	Culture-negative (n=11, 16%)
S. aureus	11 (20%)	8 (42%)	-
CNS	15 (41%)	9 (47%)	-
Streptococci[‡]	1 (3%)	6 (32%)	-
Gram-negative[^]	1 (3%)	4 (21%)	-
C. acnes	5 (14%)	2 (11%)	-
Corynebacteriae	0	2 (11%)	-
Enterococci	3 (8%)	5 (26%)	-
Anaerobic[#]	1 (3%)	8 (42%)	-

[^] *Proteus mirabilis, Enterobacter cloacae, Pseudomonas aeruginosa, Acinetobacter baumanni, Moraxella,* Klebsiella species, *Haemophilus parainfluenzae*

[#] *Peptoniphilus harei, Finegoldia magna, Clostridium paraputrificum, Lactobacillus, Clostridium perfringens, Clostridium disporicum,* Veillonella species, *Peptostreptococcus anaerobius*

[‡] *Streptococcus anginosus, Micrococcus luteus, Streptococcus oralis, Streptococcus vestibularis,* other Beta-hemolytic streptococci

Materials and Methods

Study design and population

A retrospective cohort study and a review of the literature was conducted. Institutional databases were queried to identify all patients who underwent endoprosthetic reconstruction of the lower extremities following tumor resection between 2000 and 2018 in a tertiary referral center for orthopedic oncology. Patients who subsequently developed a PJI of the tumor endoprosthesis were included. Micro-organisms isolated during the first surgical procedure for infection were recorded, as were the number of reoperations for persistent infection or secondary superinfection, antimicrobial treatment strategy and the outcome of treatment. A nested case-control study was performed to identify risk factors for treatment failure after initial DAIR. The minimum follow-up was 24 months, calculated from the moment the infection was diagnosed.

Index surgery

Tumor resection and reconstruction using a modular implant was performed in one surgical session. Proximal femur, distal femur and proximal tibia modular endoprostheses (Kotz, Howmedica/Stryker, Kalamazoo, Michigan, United States; MUTARS, Implantcast, Buxtehude, Germany) were used. A first-generation cephalosporin was administered at least 30 minutes prior to skin incision in all patients and repeated every 4 hours of surgery or in case blood loss exceeded 1.5 L. Prophylactic antibiotics were continued for 24 hours

to five days based on variables such as duration of surgery, extent of resection, wound healing and patient characteristics. Antibiotic-loaded cement, gels, and gentamicin beads were not used as local prophylaxis.

Surgical treatment for PJI

Patients underwent either surgical debridement with retention of the implant (DAIR) or prosthesis explantation as part of a two-stage revision. A DAIR procedure was the preferred initial treatment strategy in patients with either acute postoperative or late acute hematogenous infections. A thorough debridement was performed with resection of all avital tissue, mechanical cleaning of the implant with Chlorhexidine, disassembly of endoprosthetic parts, iodine pulse lavage and exchange of polyethylene and mobile parts, whenever possible. During surgery, at least five Prosthetic tissue samples were obtained for culture. Gentamicin sponges were used at surgeons discretion. Primary wound closure without a surgical drain was pursued. A primary two stage procedure was considered in patients with a chronic or low-grade PJI, a sinus tract or septic loosening of the implant. Following explantation in two stage procedures, re-implantation was considered if the inflammatory parameters normalized after a minimum of six weeks of antibiotic treatment and two weeks without antibiotics. Temporary use of gentamicin beads and spacers was considered in case of large dead spaces. Empiric antibiotic treatment was started immediately after surgical debridement and consisted of intravenous flucloxacillin and gentamicin. For patients treated with DAIR, rifampicin was added to empiric antibiotic treatment for five postoperative days, starting immediately postoperative. Rifampicin was discontinued earlier if cultures revealed Gram-negative bacteria or enterococci. Antibiotic treatment was switched to targeted therapy for at least six weeks based on antibiotic sensitivity of cultured micro-organisms. The decision to discontinue targeted therapy was made based on clinical response and inflamatory parameters. All patients were regularly discussed in a multidisciplinary team meeting (orthopedic surgeon, infectious disease physician and microbiologist attending). The decision to treat with (repeated) DAIR, two-stage exchange, amputation or chronic suppressive antibiotic treatment was guided by the expected risk of treatment failure and survival, quality of life and patient preference.

Definitions

Prosthetic joint infection was defined as presence of one or more of the following criteria: presence of pus around the prosthesis, a sinus tract communicating with the prosthesis, at least two positive intraoperative cultures with the same microorganism or one positive culture with a virulent micro-organism. Infection within six weeks was defined as an acute infection. Infection after six weeks but before three months was considered an early chronic infection. Infection after three months was considered chronic infection. Cure

was defined as an endoprosthesis in situ at the time of the latest follow-up, no draining fistula and no antibiotic therapy. Patients were considered functionally cured when an endoprosthesis was in situ at the time of the latest follow-up *with or without* chronic suppressive antibiotic therapy or a draining fistula. Implant removal or amputation were defined as treatment failure.

Statistics

Descriptive statistics were used for baseline clinical characteristics, cultured micro-organisms and clinical outcome. A nested case-control design was employed to determine which explanatory variables influenced treatment failure after initial DAIR. Logistic regression was used to compare risk factors between patients with and without failure. Results were reported as odds ratios (OR) with 95% confidence intervals (95%CI). Statistical analyses were performed using SPSS Statistics (version 25).

Results

337 patients with endoprosthetic tumor reconstruction surgery of the lower extremities were identified. Median follow-up following the index procedure was 9.5 years (95%CI=6.2-12.8). Of them, 67 (20%) patients developed a PJI. Baseline characteristics are summarized in table 1. The median age at reconstruction surgery was 52 years (IQR 23 to 65 years). Median reconstruction length was 17cm (IQR 14-22). Prosthetic joint infection (n=67) was diagnosed at a median of 1.4 months following the last surgical procedure preceding infection (IQR 0.6-7.8 months). Fifty-five (82%) patients were primarily treated with DAIR, ten (15%) patients with a two-stage procedure and two (3%) patients with direct amputation. Median follow-up after surgical debridement was 3.8 years (95%CI=2.0-5.5) (figure 1). The causative micro-organisms are summarized in Table 2. Staphylococci were the predominant causative microorganisms (*Staphylococcus aureus* 28%, Coagulase-negative staphylococci (CNS), 36%), followed by anaerobic bacteria (15%), enterococci (12%), streptococci (10%) and *Cutibacterium acnes* (10%). Eighteen (27%) patients had a polymicrobial infection. Of them, 13 patients had a polymicrobial infection with more than two micro-organisms. Nine patients (13%) remained culture negative.

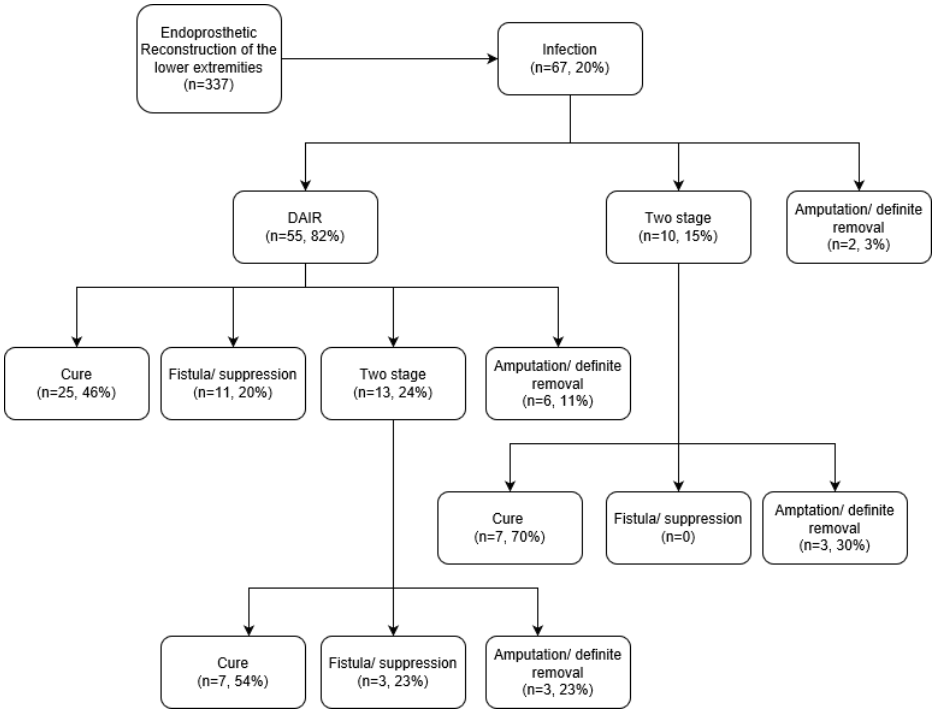


Figure 1. Outcome of 67 patients with tumour prostheses PJI of the lower extremities.

Of the 55 patients primarily treated with DAIR, a median of 2 debridements per patient was needed. Each subsequent DAIR procedure had a functional cure rate between 32 and 50% (figure 2). Thirty-six patients (65%) were functionally cured at final follow up. Of these 36 patients, 11 (31%) patients received chronic suppressive antibiotic treatment. Of these eleven patients, none had clinical signs of active infection or needed further surgical treatment at the time of latest follow-up. The decision to continue suppressive antibiotic treatment or to leave a fistula untreated was based on uncertainty of complete surgical eradication of the biofilm, patient life expectancy or patient reluctancy towards additional surgery. Of the patients with failure after one or more DAIR procedures, thirteen (24%) proceeded with a two-stage exchange of the endoprosthesis, six patients (11%) proceeded with an amputation or Girdlestone procedure. Of thirteen patients with a two-stage procedure after failed DAIR, the secondary implant could be retained in ten patients (77%) with complete cure in seven and functional cure in three patients. Three patients (23%) eventually needed amputation or a Girdlestone procedure (Figure 1).

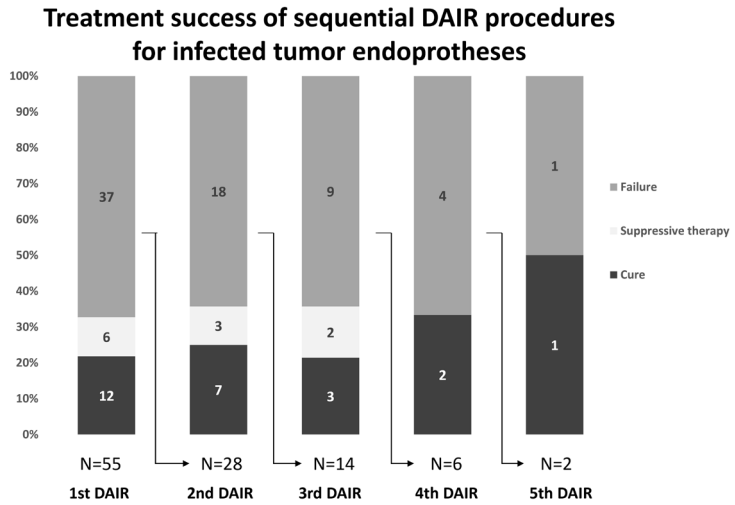


Figure 2. Outcome of sequential DAIR procedures for tumor endoprostheses PJI.

Patients in the second DAIR group consists of patients with a failure after the first DAIR who were subsequently treated with a second DAIR.

In ten patients primarily treated with a two-stage procedure, three patients (30%) did not need additional surgery, while 5 patients (50%) needed two to five extra debridements between implant removal and reimplantation. A mean of 3.1 debridements were performed per patient including reimplantation surgery. Eventually, seven patients were completely cured (70%) while three patients needed an amputation or a Girdlestone procedure (30%).

To evaluate risk factors for failure, a nested case-control was performed for the 55 patients initially treated with DAIR (Table 3). Chemotherapy (OR=3.1,95%CI=1.0-9.3, p=.05) and erythrocyte sedimentation rate (ESR) >50mm/hour at diagnosis (OR=4.5,95%CI=1.3-15.4, p=.02) were significantly associated with treatment failure. A silver coating on the prosthesis and a history of less than two revisions prior to the onset of an infected endoprosthesis showed a trend towards improved success rates after DAIR (OR=4.0,95%CI=0.8-19.8 and OR=3.6,95%CI=0.9-15.2, respectively). Time from last procedure to surgical debridement (OR=1.0,95%CI=0.9-1.0), resection length (OR=0.9,95%CI=0.99-1.01), leukocyte count at diagnosis (OR=1.0, 95%CI 0.9-1.2) and C-reactive protein at diagnosis (OR=1.0,95%CI=0.99-1.00) were not associated with treatment failure.

Table 3. Risk factors for treatment failure[$]

	Success (n = 25 cases)	Failure (n = 30 controls)	OR	95% CI	p-value
Male gender	15	20	0.75	0.25-2.26	0.61
Age (mean)	47	52	0.99	0.97-1.02	0.51
ASA (mean)	2.11	2.24	1.00	0.40-2.48	1.00
Cemented fixation	6	1	1.28	0.42-3.83	0.68
Non-silver coated implant	10	14	4.00	0.81-19.82	0.09
Gentamicin mats	5	6	1.00	0.27-3.77	1.00
Secondary infection			2.03	0.69-6.02	0.20
Revisions prior to infection (>1)	9	16	3.67	0.88-15.25	0.07
Reconstruction length: (mm)	203	195	1.00	0.99-1.01	0.75
Chemotherapy	9	19	3.07	1.02-9.26	0.05
Radiotherapy	1	1	0.81	0.18-3.62	0.78
Leukocyte count at diagnosis (mean)	9.96	12.12	1.04	0.92-1.17	0.53
CRP at diagnosis (mean)	124	70	1.00	0.99-1.00	0.26
ESR >50 at diagnosis (mean)	8/22	18/28	4.50	1.31-15.42	0.02
Polymicrobial infection	8/25	9/29	1.05	0.33-3.31	0.94
Gram negative infection	4/25	2/29	0.39	0.07-2.33	0.30

In this analysis, patients with successful outcome are regarded as cases, patients with a failure as controls. Dichotomous variables are presented as number of patients, from continuous variables, mean is shown. Univariate logistic regression analysis was used for continuous variables, Odds Ratios were calculated for dichotomous variables.

[$] In 55 patients primarily treated with DAIR

Discussion

Long term risk of infection

Twenty percent of our patients developed a PJI, which is high compared to literature (range 9-15%, table 4). However, most studies report the risk of infection during the first months after implantation rather than the life-long risk. Also, studies report the incidence of implant removal for infection rather than the true incidence of PJI. The follow-up after index surgery in our study was long. Many infections (49%) in our cohort occurred after revision procedures for mechanical complications. In a study on long-term outcomes of endoprosthetic reconstruction of tumor defects, Grimer et al. reported that 21 (9%) patients developed a PJI following the primary procedure, while 39 (14%) patients developed a PJI after successive revision procedures. They reported that the risk of PJI persists during follow-up, at a mean of 1% per year [7]. The high risk of secondary infection following revision surgery for mechanical complications stresses the importance of fixation and durability of implant designs.

Table 4. Outcome of DAIR stratified for location of infection.

Localization	Treatment success (%)
Proximal femur	6/18 (33)
Distal femur	13/27 (48)
Femur*	3/4 (75)
Proximal tibia	3/6 (50)

*Total femoral and intercalary reconstruction

Surgical treatment strategy

The cure rate in this study (65%) is comparable to other studies on tumor endoprostheses PJI (45-93%, table 2) and studies that report outcome for conventional PJI (on average 60%)[6]. However, as a result of heterogeneity of definitions of treatment success and length of follow up, outcomes are difficult to compare. We observed a higher mean number of operations in patients initially planned for two-stage revision (3.1) compared to DAIR (1.9), which can be attributed to the scheduled reimplantation. Our results show that DAIR was successful in 65% of the patients treated with one or more DAIR. A two-stage procedure could be prevented in these patients. On the other hand, 19 patients treated with two or more debridements, with associated hospital admissions and long-term antibiotic therapy, had to proceed to a two-stage procedure, amputation or definite removal of the implant. Although numbers were limited, the chance of eradicating the infection was 32-50% after each subsequent DAIR. Literature shows conflicting evidence concerning the outcome of multiple DAIR procedures in conventional arthroplasty. Some authors identified the number of sequential DAIR procedures as an independent risk factor for treatment failure [8-10]. However, other studies reported favorable outcomes of sequential DAIR [11-13].

Risk factors for treatment failure

The identification of risk factors for failure of successive DAIR procedures may guide in decision-making between repeat DAIR and a one or two-stage revision. Our study shows that chemotherapy is associated with inferior outcome in patients treated with DAIR. This might be explained by a deficient innate and/or adaptive immune response secondary to chemotherapy and/or the effects of chemotherapy on vascularization[14]. Baseline ESR >50mm/hour was also significantly associated with inferior outcome after DAIR. This might be explained by the fact that the ESR is a marker of chronic infection which may lead to inferior outcome. Other authors also identified elevated ESR as an independent risk factor for infection treatment failure after conventional hip or knee arthroplasty [13].

In two previous studies, treatment outcome after DAIR tended to be more successful with silver coated implants[15,16]. Our numbers were too low to draw conclusions. However, it seems

reasonable to continue the use of silver coated implants, although larger randomized controlled trials are needed to address this issue. Other treatment strategies, such as iodine coatings, may have added value but was not used in our cohort. In our study, the length of reconstruction was not associated with treatment outcome. Other factors, such as the quality of soft tissue coverage, may be of more importance. Unfortunately, these factors are difficult to quantify.

Based on the data presented in our study, performing one or more DAIR procedures in patients without risk factors for treatment failure seems to be a reasonable treatment strategy. When risk factors for failure are present, like a chronic PJI or recent chemotherapy, a one- or two-stage procedure, should be considered. Although the numbers are limited, two stage replacement as a salvage procedure after successively failed DAIR procedures showed reasonable success rates, justifying the choice for a step-up approach with initial DAIR. A major disadvantage of two-stage revision is the loss of bone stock complicating any future reconstruction. Furthermore, failed primary two-stage procedures usually do not leave any limbsalvaging options.

Epidemiology of micro-organisms

Most hip and knee infections were caused by *Staphylococcus aureus* (20%) and coagulase-negative staphylococci (41%), which is comparable to the literature regarding conventional PJI[17]. The proportion of PJI caused by polymicrobial flora (30%) including numerous anaerobic bacteria (42%) in our cohort is higher than wat is usually reported. Tande et al. reported 14% polymicrobial and 4% anaerobic bacteria on 1979 patients with conventional hip or knee PJI. A larger wound area after tumor reconstruction surgery and reduced local immunity may explain the higher proportion of polymicrobial infections in these patients.

Prophylactic antibiotic strategy

The preoperative antibiotic prophylaxis strategy is determined by many factors including local epidemiology, local resistance patterns, pharmacokinetic profile, bactericidal activity, cost and safety. Antibiotic stewardship bundles during surgery may further reduce the risk of transmission of bacteria to the surface of the implant. Allegedly, cefazolin prophylaxis did not prevent many *S. aureus* and streptococcal infections in this study, together counting for a third of cases in our cohort. Poor penetration of systemic antibiotics in the dead space after tumor resection may have played a role here. Local prophylactic antibiotic treatment with gentamicin beads, cement, gels, and sponges is often used to achieve high local concentrations without systemic toxicity but there is no solid evidence to support this [13]. There is even a risk that bacteria can adhere to local gentamicin beads causing secondary infections. Despite the low level of evidence, application of local antibiotics to spacers which are inserted in infected wound areas after removal of the implant and debridement seems rational.

The high percentage of polymicrobial flora in this cohort may raise the question of whether a broader spectrum of antibiotic prophylaxis is needed. The use of prophylactic cefazolin could not prevent that 30% of PJIs were caused by *S. aureus* and streptococci, possibly related to reduced local concentration in large woundbeds and other surgery-related factors. Larger observational studies are needed to define which specific patient groups are most likely to develop anaerobic and/or Gram-negative infections and who may benefit of prophylactic antibiotics with a more extended spectrum. The PARITY cohort may contribute answering this question[18].

To conclude, this study shows that patients undergoing endoprosthetic reconstruction of the lower extremities have a high risk of PJI requiring multiple surgical interventions. A significant proportion of infections is caused by revision procedures and stresses the importance of continuous innovation of tumor endoprostheses and surgical techniques to minimize revision procedures for mechanical reasons. Performing multiple DAIR procedures is a feasible treatment option when diligent patient selection is applied. Primary two-stage showed reasonable outcomes but has major drawbacks as noted in this study. In our series, we found more polymicrobial infections compared to conventional PJI. Larger observational studies are needed to identify patient groups who may benefit of specific additional prophylactic antibiotics.

Table 5. Review of studies reporting outcome of tumour prosthesis PJI.

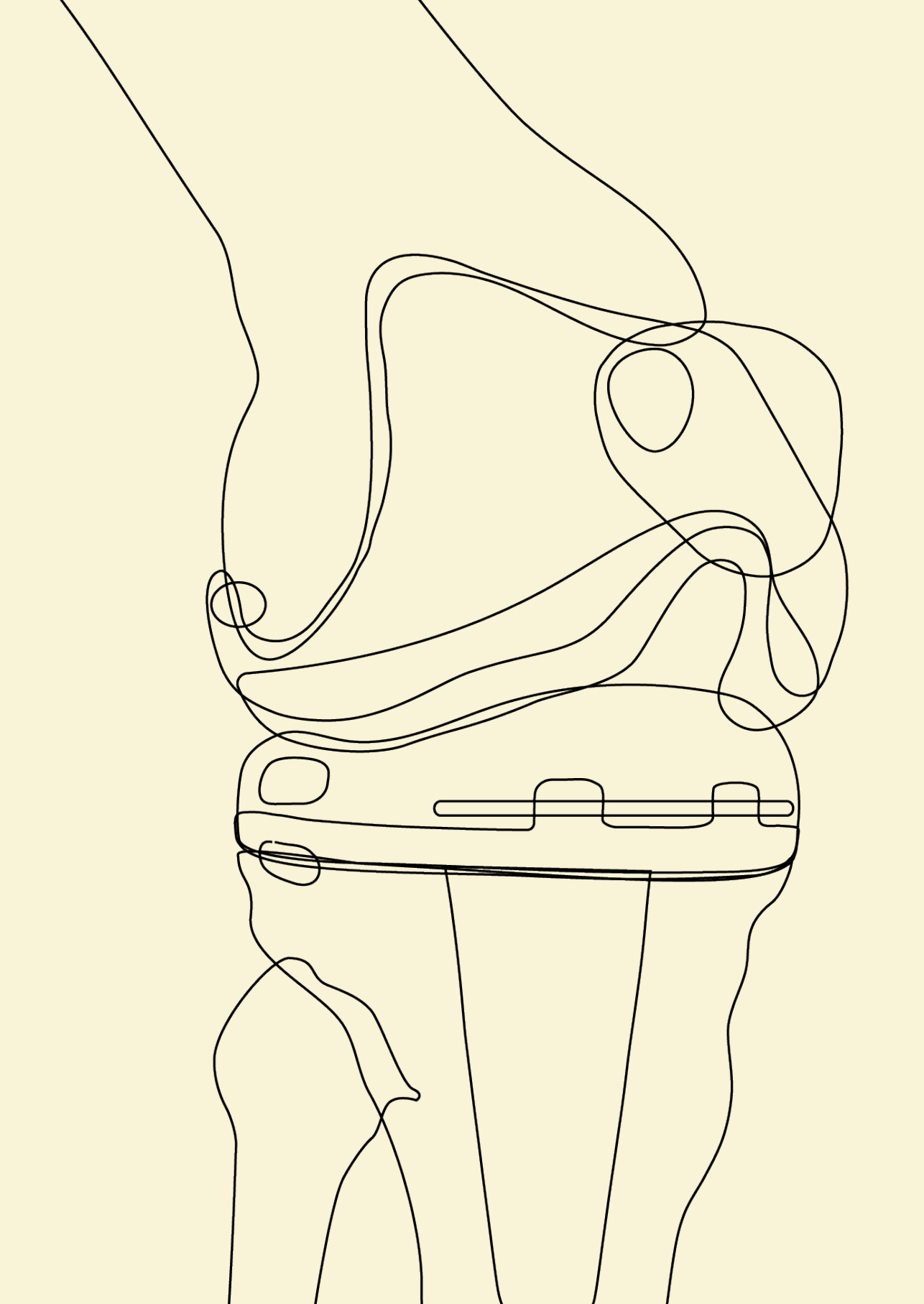
Author	Year of surgery	N patients with reconstruction surgery	Location	Implant type	Cemented (%)
Mavrogenis[19]	1983 - 2010	1161	DF 64% PT 20% PF 13% TF 3% EAK 1%	KMFTR HMRS GMRS	8
Schmolders[20]	2008 - 2014	100	PF 52% DF 30% TF 14% EXP 3% PT 1%	N/R	N/R
Pala[21]	2003 - 2010	247	DF 76%, PT 25%	GMRS	9
Bus[4]	1995 - 2010	110	DF 81% PT 19%	MUTARS	10
Jeys[5]	N/R	1240	DF 37% PF 21% PT 20% HUM 14% PEL 4% FD 3% TF 1%	N/R	N/R
De Gori[22]	2001 - 2014	87	PF 46% DF 30% PT 10% KA 9% TF 5%	MSC	60
Sigmund[23]	1982 -2017	621	DF 51% PT 31% PF 15% EAK 2% TF 1%	KMFTR HMRS GMRS MUTARS	N/R
Morii[24]	1995-2009	388	DF 59% PT 41%	HMRS Kyocera	N/R
Peel[25]	1996-2010	121	PF/DF 74% PEL 9% PT 14 HUM 3	N/R	N/R

	Silver coating (%)	Infection (%)	Primary treatment strategy	Overall cause-specific failure risk (%)	Notes
	N/R	9	12% one-stage, 83% two-stage, 5% amputation	12% at 10 years, 16% at 20 years	Higher survival rate for uncemented implants; no influence of adjuvant treatments.
	100	10	40% implant removal, 20% two-stage, 30% DAIR, 10% amputation	7	50% of patients with an infection underwent no further reconstruction after implant removal.
	N/R	N/R	25% one-stage, 75% two-stage	9	87% of patients with an infection had a successful revision.
	3	14	N/R	9	33% of infected implants were retained.
	N/R	11	43% two-stage, 31% amputation, 24% one-stage, 1% implant removal	11	Patients treated over a 37-year period. Infection risk since 1996 dropped to 4%. Radiation therapy increased the risk of infection.
	14	12	67% two-stage, 33% one-stage	10	Patients treated for non-neoplastic conditions. 3% had an allograft-prosthetic composite reconstruction.
	N/R	13	73% one-stage, 19% two-stage, 5% amputation, 2% DAIR	13	In 44% of two-stage revisions, at least one well fixed stem was retained; these had a significantly higher re-infection rate (64%) than two-stage revisions in which the entire implant was removed (22%). No significant difference in re-infection rate between one- and two-stage revision procedures. No difference in re-infection risk between silver-coated and uncoated implants.
	N/R	15	N/R	55	Only total of procedures reported. Distribution of primary procedures not reported.
	N/R	14	53% DAIR 12% One-stage 24% Two-stage 12% Amputation	18	

References

1. Jeys LM, Kulkarni A, Grimer RJ, et al. Endoprosthetic reconstruction for the treatment of musculoskeletal tumors of the appendicular skeleton and pelvis. *J Bone Joint Surg Am* 2008;90(6):1265-71. doi: 10.2106/JBJS.F.01324 [published Online First: 2008/06/04]
2. Sanders PTJ, Bus MPA, Scheper H, et al. Multiflora and Gram-Negative Microorganisms Predominate in Infections Affecting Pelvic Endoprostheses Following Tumor Resection. *J Bone Joint Surg Am* 2019;101(9):797-803. doi: 10.2106/JBJS.18.00836 [published Online First: 2019/05/03]
3. Fujiwara T, Ebihara T, Kitade K, et al. Risk Factors of Periprosthetic Infection in Patients with Tumor Prostheses Following Resection for Musculoskeletal Tumor of the Lower Limb. *J Clin Med* 2020;9(10) doi: 10.3390/jcm9103133 [published Online First: 2020/10/02]
4. Bus MP, van de Sande MA, Fiocco M, et al. What Are the Long-term Results of MUTARS® Modular Endoprostheses for Reconstruction of Tumor Resection of the Distal Femur and Proximal Tibia? *Clin Orthop Relat Res* 2015 doi: 10.1007/s11999-015-4644-8
5. Jeys LM, Grimer RJ, Carter SR, et al. Periprosthetic infection in patients treated for an orthopaedic oncological condition. *J Bone Joint Surg Am* 2005;87(4):842-9. doi: 10.2106/JBJS.C.01222 [published Online First: 2005/04/05]
6. Tande AJ, Patel R. Prosthetic joint infection. *Clin Microbiol Rev* 2014;27(2):302-45. doi: 10.1128/CMR.00111-13 [published Online First: 2014/04/04]
7. Grimer RJ, Aydin BK, Wafa H, et al. Very long-term outcomes after endoprosthetic replacement for malignant tumours of bone. *Bone Joint J* 2016;98-B(6):857-64. doi: 10.1302/0301-620X.98B6.37417 [published Online First: 2016/05/29]
8. Vilchez F, Martinez-Pastor JC, Garcia-Ramiro S, et al. Outcome and predictors of treatment failure in early post-surgical prosthetic joint infections due to Staphylococcus aureus treated with debridement. *Clin Microbiol Infect* 2011;17(3):439-44. doi: 10.1111/j.1469-0691.2010.03244.x [published Online First: 2010/04/24]
9. Deirmengian C, Greenbaum J, Stern J, et al. Open debridement of acute gram-positive infections after total knee arthroplasty. *Clin Orthop Relat Res* 2003(416):129-34. doi: 10.1097/01.blo.0000092996.90435.35 [published Online First: 2003/12/04]
10. Martel-Laferriere V, Laflamme P, Ghannoum M, et al. Treatment of prosthetic joint infections: validation of a surgical algorithm and proposal of a simplified alternative. *J Arthroplasty* 2013;28(3):395-400. doi: 10.1016/j.arth.2012.06.034 [published Online First: 2012/11/16]
11. Mont MA, Waldman B, Banerjee C, et al. Multiple irrigation, debridement, and retention of components in infected total knee arthroplasty. *J Arthroplasty* 1997;12(4):426-33. doi: 10.1016/s0883-5403(97)90199-6 [published Online First: 1997/06/01]
12. Peel TN, Cheng AC, Choong PF, et al. Early onset prosthetic hip and knee joint infection: treatment and outcomes in Victoria, Australia. *J Hosp Infect* 2012;82(4):248-53. doi: 10.1016/j.jhin.2012.09.005 [published Online First: 2012/10/23]
13. Kuiper JW, Vos SJ, Saouti R, et al. Prosthetic joint-associated infections treated with DAIR (debridement, antibiotics, irrigation, and retention): analysis of risk factors and local antibiotic carriers in 91 patients. *Acta Orthop* 2013;84(4):380-6. doi: 10.3109/17453674.2013.823589 [published Online First: 2013/07/16]

14. Cameron AC, Touyz RM, Lang NN. Vascular Complications of Cancer Chemotherapy. *Can J Cardiol* 2016;32(7):852-62. doi: 10.1016/j.cjca.2015.12.023 [published Online First: 2016/03/13]
15. Schmidt-Braekling T, Streitbuerger A, Gosheger G, et al. Silver-coated megaprostheses: review of the literature. *Eur J Orthop Surg Traumatol* 2017;27(4):483-89. doi: 10.1007/s00590-017-1933-9 [published Online First: 2017/03/08]
16. Wafa H, Grimer RJ, Reddy K, et al. Retrospective evaluation of the incidence of early periprosthetic infection with silver-treated endoprostheses in high-risk patients: case-control study. *Bone Joint J* 2015;97-B(2):252-7. doi: 10.1302/0301-620X.97B2.34554 [published Online First: 2015/01/30]
17. Manning L, Metcalf S, Clark B, et al. Clinical Characteristics, Etiology, and Initial Management Strategy of Newly Diagnosed Periprosthetic Joint Infection: A Multicenter, Prospective Observational Cohort Study of 783 Patients. *Open Forum Infect Dis* 2020;7(5):ofaa068. doi: 10.1093/ofid/ofaa068 [published Online First: 2020/05/21]
18. Prophylactic Antibiotic Regimens in Tumor Surgery I, Ghert M, Schneider P, et al. Comparison of Prophylactic Intravenous Antibiotic Regimens After Endoprosthetic Reconstruction for Lower Extremity Bone Tumors: A Randomized Clinical Trial. *JAMA Oncol* 2022;8(3):345-53. doi: 10.1001/jamaoncol.2021.6628 [published Online First: 2022/01/07]
19. Mavrogenis AF, Pala E, Angelini A, et al. Infected Prostheses after Lower-Extremity Bone Tumor Resection: Clinical Outcomes of 100 Patients. *Surg Infect (Larchmt)* 2015;16(3):267-75. doi: 10.1089/sur.2014.085 [published Online First: 2015/03/27]
20. Schmolders J, Koob S, Schepers P, et al. Lower limb reconstruction in tumor patients using modular silver-coated megaprostheses with regard to perimegaprosthetic joint infection: a case series, including 100 patients and review of the literature. *Archives of orthopaedic and trauma surgery* 2017;137(2):149-53. doi: 10.1007/s00402-016-2584-8 [published Online First: 2016/10/27]
21. Pala E, Trovarelli G, Calabro T, et al. Survival of Modern Knee Tumor Megaprostheses: Failures, Functional Results, and a Comparative Statistical Analysis. *Clinical orthopaedics and related research* 2014 doi: 10.1007/s11999-014-3699-2 [published Online First: 2014/05/31]
22. De Gori M, Scoccianti G, Frenos F, et al. Modular Endoprostheses for Nonneoplastic Conditions: Midterm Complications and Survival. *Biomed Res Int* 2016;2016:2606521. doi: 10.1155/2016/2606521 [published Online First: 2017/01/05]
23. Sigmund IK, Gamper J, Weber C, et al. Efficacy of different revision procedures for infected megaprostheses in musculoskeletal tumour surgery of the lower limb. *PLoS One* 2018;13(7):e0200304. doi: 10.1371/journal.pone.0200304 [published Online First: 2018/07/06]
24. Morii T, Yabe H, Morioka H, et al. Postoperative deep infection in tumor endoprosthesis reconstruction around the knee. *J Orthop Sci* 2010;15(3):331-9. doi: 10.1007/s00776-010-1467-z [published Online First: 2010/06/19]
25. Peel T, May D, Buising K, et al. Infective complications following tumour endoprosthesis surgery for bone and soft tissue tumours. *Eur J Surg Oncol* 2014;40(9):1087-94. doi: 10.1016/j.ejso.2014.02.241 [published Online First: 2014/03/25]



Part III. New antimicrobial strategies for PJI

CHAPTER 6

Effectiveness of different antimicrobial strategies for staphylococcal prosthetic joint infection: results from a large prospective registry-based cohort study

Henk Scheper, Robert J.P. van der Wal, Rachid Mahdad, Stefan Keizer, Nathalie M. Delfos, Joris C.T. van der Lugt, Karin Ellen Veldkamp, Peter A. Nolte, Masja Leendertse, Luc B.S. Gelinck, Femke P.N. Mollema, Emile F. Schippers, Hanke G. Wattel-Louis, Leo G. Visser, Rob G.H.H. Nelissen, Mark G.J. de Boer

Open Forum Infect Dis 2022, 9(10): ofac474

Abstract

Background

Treatment of staphylococcal prosthetic joint infection (PJI) usually consists of surgical debridement and prolonged rifampicin combination therapy. Tailored antimicrobial treatment alternatives are needed due to frequent side effects and drug-drug interactions with rifampicin combination therapy. We aimed to assess the effectiveness of several alternative antibiotic strategies in patients with staphylococcal PJI.

Methods

In this prospective, multicenter registry-based study, all consecutive patients with a staphylococcal PJI , treated with DAIR or one-stage revision surgery between January 1st, 2015 and November 3rd, 2020, were included. Patients were treated with a long-term rifampicin combination strategy (in two centers) or a short-term rifampicin combination strategy (in three centers). Antimicrobial treatment strategies in these centers were defined before the start of the registry.. Patients were stratified in different groups, depending on the used antimicrobial strategy. Cox proportional hazards models were used to compare outcome between the groups.

Results

Two hundred patients were included and, based on the antimicrobial treatment, stratified in one long-term rifampicin group (traditional rifampicin combination therapy) or one of the three short-term rifampicin groups (clindamycin or flucloxacillin or vancomycin monotherapy, including rifampicin for only five postoperative days). Adjusted hazard ratios for failure for patients treated with either flucloxacillin or clindamycin were almost equal to patients treated with long-term rifampicin combination therapy (aHR 1.21, 95%CI 0.34-4.40).

Conclusions

A short-term rifampicin strategy with either clindamycin or flucloxacillin and only five days of rifampicin was found to be as effective as traditional long-term rifampicin combination therapy. A randomized controlled trial is needed to further address efficacy and safety of alternative treatment strategies for staphylococcal PJI.

Introduction

A prosthetic joint infection (PJI) is a serious complication occurring in 1-2% of patients with a joint arthroplasty resulting in prolonged hospitalization, impaired mobility and long-term antibiotic treatment[1,2]. Most PJIs are caused by staphylococci most of which most are highly susceptible for rifampicin, clindamycin and flucloxacillin in The Netherlands (MRSA is virtually absent in our region)[3]. Treatment of acute PJI consists of thorough surgical debridement combined with antimicrobial therapy. Adequate debridement is of utmost importance as the biofilm that has been formed on the surface of the implant needs to be removed as much as possible to enable cure. Antimicrobial therapy consists of intravenous antibiotics for up to two weeks followed by targeted oral antimicrobial therapy[4].. For staphylococcal PJI, rifampicin and fluoroquinolone combination therapy is advocated by most national guidelines. However, its use is hampered in practice by drug-drug interactions and significant side effects underscoring the need for safe and effective alternative antimicrobial regimens for PJI[5,6]. Further, the evidence for this antibiotic strategy in clinical studies for staphylococcal PJI is lacking[7,8]. Also, studies investigating tailored alternative strategies for rifampicin combination treatment are scarce[9,10]. In 2015, a regional group of specialized centers for PJI decided to intensify collaboration and harmonized their local protocols for antimicrobial and surgical treatment. In those centers, several different antibiotic strategies, which were consistent within a center, were accepted as routine care to treat staphylococcal PJI after DAIR (Debridement, Antibiotics and Implant Retention) or 1-stage exchange: a long-term rifampicin strategy (consisting of 12 weeks rifampicin combination therapy) and several short-term rifampicin strategies, consisting of only five days of rifampicin combination treatment, started immediately postoperative, followed by clindamycin, flucloxacillin or vancomycin monotherapy. The collaborating centers initiated a web-based quality registry to evaluate the outcome of PJI after implementation of this protocol. The main objective of this prospective study is to compare the effectiveness of long-term rifampicin combination treatment with several short-term rifampicin antimicrobial strategies for the treatment of staphylococcal PJI.

Methods

Study Design

This multicenter, prospective registry-based cohort study was conducted as part of the Prosthesis Protect Project (PPP). This prospective quality registry comprised five regional hospitals in the south-western area in the Netherlands that coordinated treatment for patients with PJI. A treatment protocol for PJI was written by all collaborators prior to data collection in the database. As for registration of data, all treatment decisions and deviations from the protocol were discussed during weekly multidisciplinary meetings (MDT) with orthopedic surgeons, infectious diseases physicians and/or clinical microbiologists. Data were collected in a secured online database and double-checked by the coordinating investigator; discrepancies were resolved by consensus. The study was approved by the institutional review board and conducted according to Dutch law and regulations regarding medical research. All patients with PJI were informed by their treating physician about the quality registry and were included in the database unless they opted out.

Patient Consent Statement

The study was approved by the institutional review board of Leiden University Medical Center with a waiver of written informed consent and conducted according to Dutch law and regulations regarding medical research. All patients with PJI were informed by their treating physician about the quality registry and were included in the database unless they opted out.

Data collection and treatment protocol

For the current study, all patients aged 18 years or older with staphylococcal PJI treated with DAIR or one-stage exchange between January 1st, 2015 and November 3rd, 2020 were eligible for inclusion. Only these surgical strategies were included because the focus of this study is on the role of antimicrobial therapy in the context of retained or newly inserted implants in an infected area. Patients with polymicrobial PJI including staphylococci were also included. Patients with infected megaprostheses (e.g., after tumor resections) were excluded. PJI was defined in compliance with the Infectious Diseases Society of America (IDSA) guideline on PJI[4]. The diagnostic and debridement procedure was completely standardized between the centers (see Supplemental Table 1). Patients with acute PJI were treated with DAIR. One-stage exchange was performed in patients with chronic PJI. Empiric antimicrobial therapy for PJI was started after intraoperative cultures were taken.

Definitions

PJI was defined as acute PJI when diagnosed within 3 weeks after onset of clinical symptoms or within 3 weeks after implantation or last revision of the implant. All other PJIs were defined as chronic PJI. For the current study, patients were also stratified in early acute PJI

(within three weeks after arthroplasty or revision), early chronic PJI (between three weeks and three months after arthroplasty or revision), late chronic PJI (more than three months after arthroplasty or revision, caused by low-virulent micro-organisms) and late acute PJI (more than three months after arthroplasty or revision, caused by virulent micro-organisms (e.g., *S. aureus*). Cure was defined as absence of clinical symptoms of infection and a retained implant during at least 12 months follow-up after antibiotic therapy was terminated AND if failure criteria were not met. Failure was defined as either (i) chronic suppressive antibiotic therapy with implant retention, (ii) a second debridement after finishing antibiotic therapy, (iii) the need for more than two debridements, (iv) removal of the implant or (v) PJI-related death. Secondary failures with other micro-organisms were also counted as failure.

Table 1. Overview of treatment schedules in the protocol for both the long-term and the short-term rifampicin strategies.

Protocol strategies	Long-term rifampicin strategy	Short-term rifampicin strategy
Antibiotic groups	rifampicin-based*	flucloxacillin-based$ clindamycin-based& vancomycin-based@
1st phase: intravenous antibiotics	flucloxacillin or vancomycin^	flucloxacillin or vancomycin^
2nd phase: targeted antibiotics	rifampicin + levofloxacin (or other antibiotics#)	flucloxacillin or clindamycin or vancomycin (or other antibiotics#)
Timing of start rifampicin	when wound is dry and antibiotic sensitivity is known	immediately postoperative after DAIR
Dose of rifampicin	300mg twice daily	600mg twice daily
Treatment duration with rifampicin	12 weeks	5 days
Total antibiotic treatment duration	12 weeks	6-12 weeks**

*Rifampicin-based: survival after DAIR >2weeks and rifampicin use for >14 days and rifampicin use for >50% of time.

$Flucloxacillin-based: survival after DAIR >2weeks and rifampicin use ≤14 days and (flucloxacillin for >50% of time or intravenous flucloxacillin for >4 weeks of time) and flucloxacillin use longer than vancomycin use (if both were used)

&Clindamycin-based: survival after DAIR >2weeks and rifampicin use ≤14 days and clindamycin use >50% of time and intravenous flucloxacillin/vancomycin < 4 weeks of time

@Vancomycin-based: survival after DAIR >2weeks and rifampicin use ≤14 days and vancomycin for >50% van time or intravenous vancomycin for >4 weeks of time and rifampicin use ≤14days and vancomycin used longer than flucloxacillin (if both were used)

#Other antibiotics: all treatment schedules that did not fit in strategies that were defined above. For long-term rifampicin combination therapy, other strategies ware accepted as long as rifampicin was combined with a second antibiotic.

^Vancomycin was given for flucloxacillin-resistant Coagulase-negative staphylococci and certain polymicrobial co-infections (e.g., corynebacteriae, enterococci). MRSA is very rare in the Netherlands (there are no patients with MRSA PJI in this cohort).

** For short-term rifampicin strategies, exact duration of antibiotics was decided in multidisciplinary team meeting. Total duration of antibiotic treatment was calculated until end of treatment or until the day of failure.

Empiric and targeted antimicrobial strategy

In all centers, empiric antibiotic therapy after surgery consisted of flucloxacillin (6gram i.v./24hrs) plus an aminoglycoside until targeted therapy could be started, based on cultures and antibiotic sensitivity. The timing of the iv-to-oral switch was after one to two weeks. In three centers, rifampicin (600mg twice daily) was added to empiric treatment for only five postoperative days, starting immediately postoperative[11]. For the purpose of this study, this was defined as a 'short-term rifampicin' strategy. In this strategy, oral targeted therapy consisted of clindamycin (600mg three times daily) or flucloxacillin (1000mg 4 or 5 times a day), dependent on susceptibility, documented allergy or intolerance. For flucloxacillin, an adequate absorption test was required, defined as a serum flucloxacillin concentration that increased at least 10mg/L after an oral loading dose of 1000mg[12]. If preferred treatment options were not available, alternative antibiotics were chosen, depending on the antibiogram. Total treatment duration was between six and twelve weeks, based on the clinical and biochemical response, such as to be decided by the MDT.

In two other centers patients with staphylococcal PJI were treated with long-term rifampicin combination therapy, the accepted standard-of-care treatment for staphylococcal PJI after DAIR[4]. Oral rifampicin (300mg twice daily) was first added to intravenous treatment once antibiotic susceptibility for rifampicin was confirmed and the postoperative wound was dry. After two weeks, it was combined with levofloxacin for a fixed treatment duration of 12 weeks. The differences in timing and duration of rifampicin between the two clusters were defined in advance in the protocol, using hospital as an instrumental variable with patients being assigned to either a long-term or one of the short-term rifampicin strategies. For the purpose of this study, patients were classified in five groups: a rifampicin-based group, a flucloxacillin-based group, a clindamycin-based group, a vancomycin-based group and a non-defined 'other antibiotics' group consisting of patients who did not meet the criteria for the first four groups (Table 1).

Statistical analysis

Clinical characteristics at baseline were summarized using descriptive statistics, stratified by antibiotic strategies. Differences between antibiotic groups were compared with Chi-square testing for categorical variables, one-way ANOVA for continuous variables and Mann-Whitney U tests for non-normally distributed continuous variables. Kaplan-Meier curves were constructed to report outcome by the different antibiotic groups. Patients were counted as failure if PJI was the direct cause of death. Patients were censored at the time of death if they died during follow up due to an event not related to PJI. A Cox proportional hazards regression model was used to investigate whether differences in outcome were associated with baseline differences between groups. Variables in the multivariate model were selected based on the univariate regression analysis. Results

are reported as hazard ratios (HR) with 95% confidence intervals (95%CIs). To prevent immortal time bias in the five antibiotic groups and to focus on the targeted treatment phase for PJI, the minimal survival time required for inclusion in the survival analysis was defined as at least 15 days after debridement. SPSS Statistics for Windows was used (IBM SPSS Statistics for Windows, Version 25.0. Armonk, NY)

Results

Of 493 patients currently registered in the database, 200 patients were included (Figure 1). Baseline clinical characteristics of the five antimicrobial strategy groups are summarized in Table 2. The proportion of *S. aureus* PJI, and bacteremia and were higher in the flucloxacillin-based group compared to the other groups ($p<0.05$). Patients in the vancomycin-based and other antibiotics group had more polymicrobial PJI, including enterococci and corynebacteriae. Follow up data are summarized in Table 3. According to the protocol, treatment duration with rifampicin was only five days in the short-term rifampicin groups. Total antimicrobial treatment duration was longer in the long-term rifampicin group (12 weeks) compared to the short-term rifampicin groups (8 weeks) (p 0.006). Four patients in the rifampicin-based group received rifampicin for only 3-6 weeks. In the flucloxacillin group, cure rate was 88% (14/16) in patients who continued with oral flucloxacillin after two weeks intravenous flucloxacillin and 74% (23/31) in patients with prolonged intravenous flucloxacillin (Table 3). In only 32% of failures, the same causative staphylococci could be cultured again.

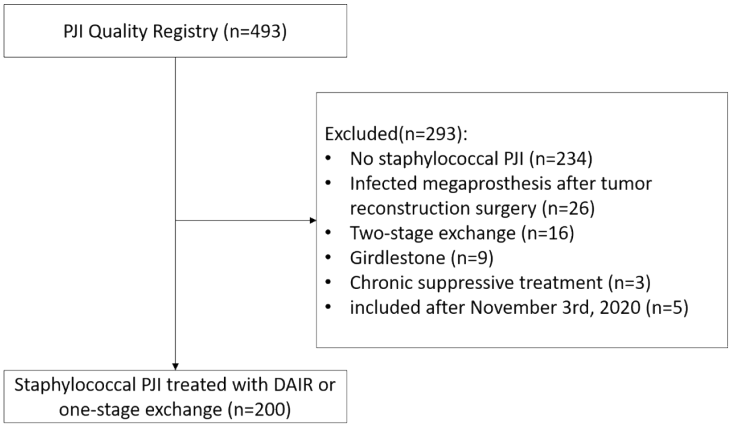


Figure 1. Flowchart of inclusion for current study.

Table 2. Baseline characteristics of all patients and after stratification for antibiotic treatment strategy.

		5 antibiotic treatment strategy groups (n= 200)[$]					
	All	**Rifampicin-based**	**Clindamycin-based**	**Flucloxacillin-based**	**Vancomycin-based**	**All other strategies***	**P value**
N patients	**200**	**23**	**56**	**47**	**26**	**48**	**-**
General characteristics							
Male sex (%)	95 (48)	11 (48)	29(52)	23 (49)	10 (39)	22 (46)	0.86
Age in years (SE mean)	70.3 (0.9)	68.8 (2.9)	67.2 (1.7)	70.1 (2.1)	72.3 (2.1)	73.6 (1.7)	0.12
Joint							
Hip	131 (66)	14 (61)	37 (66)	30 (64)	20 (77)	30 (63)	0.75
Total hip arthroplasty	109 (85)	12 (86)	32 (87)	28 (93)	15 (75)	22 (73)	0.70
Hemiarthroplasty	20 (16)	2 (14)	5 (13)	2 (7)	5 (25)	8 (27)	-
Total knee arthroplasty	63 (32)	7 (30)	17 (32)	16 (34)	6 (23)	16 (33)	0.90
Shoulder	5 (2.5)	2 (9)	1 (2)	1 (2)	0	1 (2)	-
Elbow	1 (0.5)	0	0	0	0	1 (2)	-
Previous revision	52 (26.0)	8 (35)	11 (20)	9 (19)	5 (22)	19 (40)	0.08
Previous PJI of same implant	10 (5.0)	0	1 (2)	4 (9)	0	5 (10)	0.09
Comorbidities							
Diabetes n (%)	48 (24.0)	5 (22)	12 (21)	10 (21)	9 (35)	12 (25)	0.73
Chronic kidney disease (eGFR <60ml/min)	21 (10.6)	3 (13)	4 (7)	4 (9)	4 (15)	6 (13)	0.73
Rheumatoid arthritis	13 (6.5)	3 (13)	2 (4)	3 (6)	1 (4)	4 (8)	0.57
Immunosuppressants	15 (7.5)	2 (9)	3 (5)	6 (13)	0	4 (8)	0.35
Malignancy	14 (7.0)	0	6 (11)	3 (6)	1 (4)	4 (8)	0.49
Reported smoking (n=160)	26 (13.0)	9 (39)	6 (11)	2 (4)	3 (12)	6 (13)	-
Body Mass Index (mean, SE)	30 (0.42)	28 (1.3)	30 (0.8)	29 (1.0)	30 (1.0)	30 (0.8)	0.57
Clinical Presentation							
Bacteraemia	25 (12.5)	4 (17)	4 (7)	11 (23)	0	6 (13)	0.02
Antibiotic pretreatment	31 (15.5)	3 (13)	10 (18)	7 (15)	2 (8)	9 (19)	-
Reported symptoms:							
Fever >38.3°C	40 (20.0)	5 (22)	10 (18)	16 (34)	1 (4)	8 (17)	-
Pain	107 (53.5)	11 (48)	32 (57)	31 (66)	8 (31)	24 (50)	-
Redness	94 (47.0)	5 (22)	31 (55)	21 (45)	11 (42)	26 (54)	-
Wound leakage	120 (60.0)	16 (70)	31 (55)	22 (47)	23 (89)	28 (58)	-
Fistula	4 (2.0)	0	0	3 (6)	1 (4)	1 (2)	-
Suppuration	25 (12.5)	4 (17)	5 (9)	7 (15)	3 (12)	6 (13)	-
Laboratory values							
CRP (median, range)	81 (1-585)	85 (2-313))	74 (3-443)	157 (1-585)	69 (10-342)	100 (1-491)	0.04
ESR (median, range)	49 (2-140)	53 (8-130)	41 (7-120)	53 (2-120)	46 (4-140)	58 (5-133)	-
Leukocytes (mean, SE)	11.2 (0.3)	11.1 (1.1)	11.2 (0.5)	11.9 (0.7)	10.1 (1.2)	11.1 (0.7)	0.64

Table 2. Continued

	All	5 antibiotic treatment strategy groups (n= 200)[$]					
		Rifampicin-based	Clindamycin-based	Flucloxacillin-based	Vancomycin-based	All other strategies*	P value
Causative microorganisms (n,%)							
S. aureus	120 (60)	13 (57)	35 (63)	39 (83)	8 (31)	25 (52)	0.00
Coagulase-negative staphylococci	89 (45)	11 (48)	22 (39)	9 (19)	20 (77)	27 (56)	0.00
S. epidermidis	64 (32)	5 (22)	12 (21)	7 (15)	19 (73)	21 (44)	-
S. lugdunensis	13 (7)	4 (17)	3 (5)	2 (4)	0	4 (8)	-
S. capitis	8 (4)	2 (9)	6 (11)	0	0	0	-
other CNS	8 (4)	1 (4)	4 (7)	0	1 (4)	2 (4)	-
Polymicrobial PJI	70 (36)	11 (48)	11 (20)	10 (21)	15 (58)	23 (48)	0.00
Staphylococci + streptococci	15 (8)	2 (9)	0	2 (4)	3 (12)	8 (15.1)	-
Staphylococci + Gram negatives	20 (10)	4 (17)	3 (5)	3 (6)	2 (8)	8 (17)	-
Staphylococci + *C. acnes*	5 (3)	0	2 (4)	0	1 (4)	2 (4)	-
Staphylococci + corynebacteriae	16 (8)	1 (4)	1 (2)	4 (9)	6 (23)	4 (8)	-
Staphylococci + enterococci	23 (12)	3 (13)	1 (2)	2 (4)	6 (23)	11 (23)	-
Staphylococci + anaerobic bact.	7 (4)	0	1 (2)	1 (2)	3 (12)	2 (4)	-
Classification PJI – 4 groups[#] (n,%)							
Early postoperative PJI(<3w)	94 (47)	13 (57)	22 (39)	20 (43)	19 (73)	20 (42)	0.13
Early chronic PJI(3w-3m)	53 (27)	6 (26)	19 (34)	11 (23)	5 (19)	12 (25)	0.13
Late chronic PJI(>3m)	18 (9)	1 (4)	8 (14)	2 (4)	1 (4)	6 (13)	0.06
Hematogenous PJI	35 (17)	3 (13)	7 (13)	14 (30)	1 (4)	10 (21)	0.03

[$] Exact inclusion criteria for each antibiotic subgroup are defined in Table 1. All patients in the flucloxacillin, clindamycin, vancomycin or 'other' group were also treated with five days of rifampicin starting immediately postoperative after DAIR.

 # Early postoperative PJI = PJI within 3weken of implantation or last revision. *Early chronic PJI* = PJI after 3 weeks but within 3 months after implantation or last revision. *Late chronic PJI* = PJI > 3months after implantations or last revision AND low-virulent micro-organisms. *Hematogenous PJI* = PJI >3months after last revision or implantation AND highly virulent micro-organisms (*S. aureus*, *E. Coli*, *Pseudomonas aeruginosa*, Enterococci, Streptococci, *Proteus spp*, *Klebsiella spp*, *Enterobacter*, other non-fermenters

* Amoxicillin (n=9), Amoxicillin-clavulanic acid (n=3), Levofloxacin (n=4), Linezolid (n=8), Cefuroxim (n=3), Doxycycline (n=3), Cotrimoxazole (n=10), Ciprofloxacin (n=4)

Table 3. Follow up and treatment outcome characteristics of all patients and after stratification for antibiotic treatment strategy.

	All (n= 200)	5 antibiotic treatment strategy groups[#] (n= 200)					
		Rifampicin-based*	Clindamycin-based	Flucloxacillin-based	Vancomycin-based	All other strategies	P value
N patients	**200**	**23**	**56**	**47**	**26**	**48**	**-**
Antibiotic strategy (median days, IQR)							
Duration antimicrobial treatment	57 (6-765)	94 (85-103)	56 (40-62)	41 (33-50)	55 (15-131)	53 (33-73)	0.001
Flucloxacillin i.v.	11 (0-385)	12 (2-22)	13 (8-18)	31 (18-44)	3 (0-5)	3 (0-6)	-
Flucloxacillin p.o.		-	-	33 (24-42)	-	-	-
Duration rifampicin treatment	5 (0-373)	86 (78-94)[&]	5 (5-5)	5 (4-6)	5 (4.5-5.5)	5 (4-6)	0.000
Time to start rifampicin	0 (0-11)	4 (2-6)	0 (0)	0 (0)	0 (0)	0 (0)	0.000
Surgical treatment strategy (n, %)							
DAIR	189 (94)	22 (96)	51 (91)	45 (96)	25 (96)	46 (96)	0.78
Reported head exchange hip	20/122 (16)	2/12 (17)	1/33 (3)	7/27 (26)	4/19 (21)	6/29 (21)	-
Reported liner exchange knee	37/61 (61)	5/7 (71)	13/17 (76)	7/16 (44)	3/6 (50)	9/15 (60)	-
One-stage revision procedure	11 (6)	1 (4)	5 (9)	2 (4)	1 (4)	2 (4)	-
Surgical interventions during treatment							
Re-DAIR needed	86 (43)	9 (39)	13 (23)	23 (49)	16 (62)	25 (52)	0.005
Time to re-DAIR (median days, range)	16 (3-407)	9 (3-14)	18 (3-336)	16 (5-152)	23 (10-407)	15 (5-358)	-
1 Re-DAIR in cured patients	36	6	5	12	7	6	-
2 Re-DAIRs in cured patients	6	0	3	2	1	0	-
Failure							
Failure or death due to PJI	53 (27)	3 (13)	5 (9)	10 (21)	8 (31)	27 (56)[@]	
Time to failure (days, range)	84 (6-410)	191 (103-274)	154 (85-399)	47 (20-397)	33 (21-410)	68 (6-381)	
Confirmed relapse with same staphylococci	17 (32)	1/3 (33)	3/5 (60)	3/10(30)	1/8 (13)	9 (33)	

i.v. intravenously; p.o. per os; DAIR: Debridement, Antibiotics and Implant Retention.

[#] Definitions of inclusion criteria per antibiotic subgroup are defined in Table 1

[&]All patients received at least 3 weeks of rifampicin. 4 patients received rifampicin for only 3-6 weeks

*Used antibiotics in addition to rifampicin: levofloxacin (500mg twice daily n=12), ciprofloxacin (n=2), flucloxacillin (n=3), amoxicillin (n=1), amoxicillin-clavulanic acid (n=1), cefalexin (n=1), clindamycin (n=2), vancomycin (n=1), cotrimoxazole (n=1)

[@]As defined in Table 1, this group contains all failures within 2 weeks (n=9)

The survival curves for the different antibiotic strategies are shown in Figure 2. Cure rates in the clindamycin group (91%) and the flucloxacillin group (79%) did not differ significantly from the rifampicin group (87%, p 0.20). Patients treated with vancomycin or not treated according to a predefined regimen had a worse outcome. Within the vancomycin-based group, success rates were lower for polymicrobial PJI with enterococci (p=0.02). Diabetes mellitus and duration of symptoms more than three weeks were significantly associated with failure in the univariate Cox regression model (Table 4). Late acute PJI, enterococcal PJI and bacteremia were associated with a worse outcome, although not statistically significant (Table 4 and Supplemental Figure 1). The adjusted hazard ratios for failure in the clindamycin group (HR 0.84, 95% CI 0.20-3.55), the flucloxacillin group (HR 2.21, 95% CI 0.60-8.17) or the combined clindamycin and flucloxacillin group (HR1.21, 95% CI 0.34-4.40) remained equal to the rifampicin-based group.

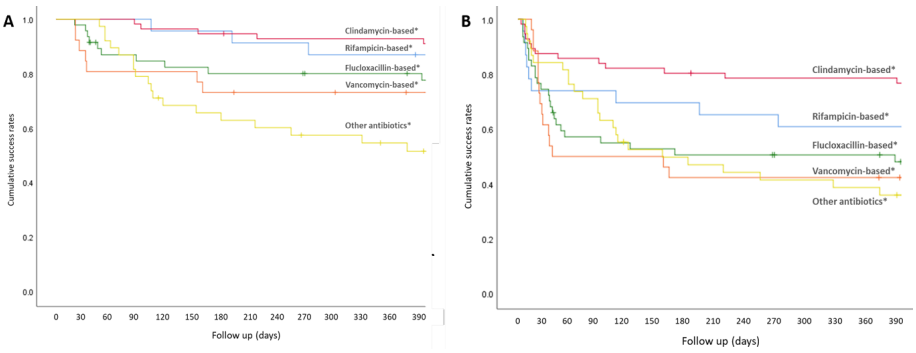


Figure 2. Survival analysis for staphylococcal PJI related to antimicrobial treatment strategy.

Figure 2A: Success rates over time for the different antibiotic groups as defined in Table 1. Figure 2B: success rates over time for the same antibiotic groups but using a narrower definition of failure in which all patient who needed a second surgery were counted as failure.

Table 4. Univariate and Multivariate Cox proportional Hazards model of clinical characteristics associated with failure.

Covariate	Univariate		Multivariate#	
	HR	95% CI	HR	95%CI
Male sex	1.35	0.74-2.46		
Revision before PJI	1.48	0.78-2.79*	1.55	0.79-3.03
Knee PJI	0.99	0.53-1.87		
Corticosteroid use	1.09	0.34-3.53		
DM	2.15	1.16-3.98*	2.12	1.14-3.42
RA	1.20	0.37-3.89		
S. aureus PJI	0.89	0.49-1.61		
Bacteraemia	1.75	0.78-3.93*	2.66	1.09-6.48
Duration of symptoms < 3weeks	0.46	0.23-0.94*	0.37	0.18-0.77
Polymicrobial PJI	0.98	0.53-1.81		
Enterococci as copathogen	1.91	0.89-4.12*	1.48	0.64-3.42
Classification PJI:				
Early postoperative	Ref.	-		
Early chronic	0.94	0.44-2.01		
Late chronic	1.05	0.36-3.08		
Late acute (hematogenous)	1.80	0.84-3.85		
Long-term rifampicin strategy center^	1.26	0.53-2.98		
Treatment strategy:				
Rifampicin-based	Ref.	-	Ref.	-
Either clindamycin- or flucloxacillin-based	1.20	0.35-4.15	1.21	0.34-4.40
Clindamycin-based	0.69	0.16-2.87	0.84	0.20-3.55
Flucloxacillin-based	1.98	0.54-7.19	2.21	0.60-8.17
Vancomycin-based	2.93	0.78-11.06	3.68	0.95-14.24
Other strategy	4.69	1.38-15.96	4.86	1.41-16.78
Exchange of liner	1.27	0.65-2.50		

#Included variables (*) in multivariate model were based on (trend to) significance in univariate model: revision before PJI, bacteremia at diagnosis, diabetes mellitus, duration of symptoms, enterococci, antimicrobial treatment strategy

^long-term rifampicin center: center where default antimicrobial strategy consisted of 12 weeks rifampicin combination therapy (see Table 1)

Discussion

There is an urgent need for alternative antimicrobial strategies for staphylococcal PJI as the current strategy with long-term rifampicin-based combination therapy is associated with significant side effects and interactions[5 6 13]. In the current study, outcome of PJI after DAIR or one-stage exchange was not statistically different between patients treated with long-term rifampicin combination therapy and patients treated with clindamycin or flucloxacillin monotherapy including only five days of rifampicin combination therapy. Moreover, treatment duration was four weeks shorter in the clindamycin-based and flucloxacillin-based groups. After correction for confounding covariates that were not evenly distributed across the groups at baseline, the outcomes in a multivariate Cox proportional hazards model did not change. Diabetes mellitus, bacteremia and a longer duration of symptoms (more than three weeks) were independent risk factors for failure.

Clindamycin monotherapy for staphylococcal PJI

Clindamycin is known to have an excellent bioavailability and penetrates well into synovial fluid and bone[14]. Reasonable outcome with clindamycin therapy for staphylococcal PJI has been incidentally published before, but this is the first study reporting the systematic use of clindamycin monotherapy[15 16]. Physicians in the short-term rifampicin strategy centers had no specific preference for either clindamycin or flucloxacillin, except that clindamycin was easier to use due to a lower pill burden. The choice for either clindamycin or flucloxacillin was completely unbiased in patients with clindamycin-resistant staphylococci or an inadequate flucloxacillin absorption test, but this was the case in only a minority of patients[12]. How should we interpret the finding that eight weeks of clindamycin-based treatment, including five initial days of rifampicin, was equivalent to twelve weeks rifampicin combination therapy and superior to flucloxacillin? Confounding by indication is the most likely explanation because in patients who needed a second debridement or who had persisting high inflammatory parameters, the iv-oral switch from flucloxacillin to clindamycin was postponed on purpose (as illustrated in Table 3 with longer i.v. treatment duration and more second DAIRs in the flucloxacillin-group). Consequently, more patients with a worse course met the criteria for the flucloxacillin-based group, leading to selection bias in favor of the clindamycin-based group. Correction for this confounding was performed by combining both groups, resulting in a cure of 85% in the combined group, which was equivalent to the rifampicin group (87%, p=0.77, Figure 3).

Flucloxacillin monotherapy for staphylococcal PJI

Clinical data regarding the use of flucloxacillin for bone and joint infections are scarce[17]. This study shows 78% success rates for staphylococcal PJI in the flucloxacillin-based group. The high success rate of 88% in the subgroup of patients treated with oral flucloxacillin

monotherapy suggests that oral flucloxacillin may be an adequate treatment strategy for staphylococcal PJI. The results in this study are congruent with an earlier small cohort study by the same authors describing reasonable outcome for staphylococcal PJI with oral flucloxacillin and short-term addition of rifampicin[11]. However, the efficacy of oral flucloxacillin for targeted treatment of staphylococcal PJI should be further assessed in a large trial. Also, a flucloxacillin absorption test is needed to identify patients with adequate oral absorption of flucloxacillin[12].

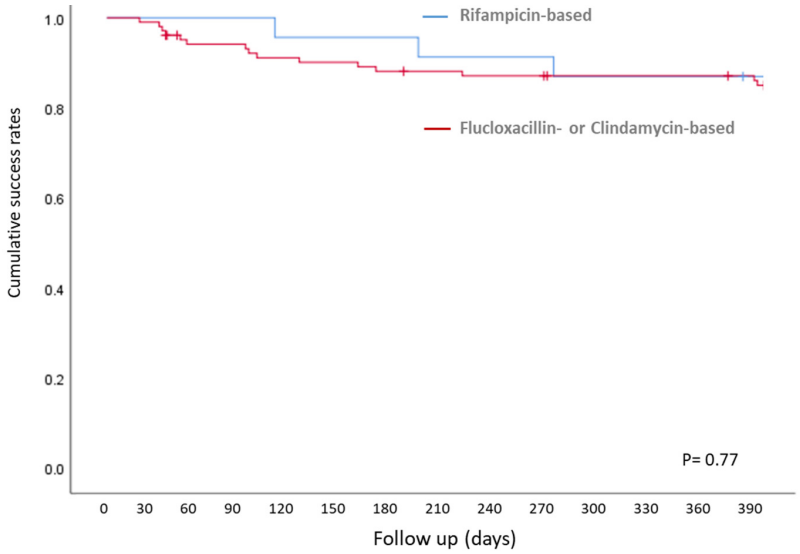


Figure 3. Survival curve after DAIR for staphylococcal PJI comparing a rifampicin-based strategy with a strategy of either flucloxacillin- or clindamycin-based treatment.

Rifampicin combination therapy for staphylococcal PJI

The effectivity of long-term rifampicin combination therapy in this study is in line with other studies reporting good outcome with this strategy[8,18]. The strength of the current study is that two different and predefined strategies between centers could be directly compared, which minimized confounding by indication between the long-term and the short-term rifampicin-based groups. However, treatment may have varied in other ways not captured by the protocol as the treatment teams between the participating centers were different. The outcome of staphylococcal PJI over time did not differ between centers with either a standard short-term or long-term rifampicin treatment strategy (Figure 4). Most surgeons in participating hospitals were educated and trained in the same program. Due to later connection of long-term rifampicin treatment centers to the registry, less patients on long-term rifampicin could be included. However, given the high cure rate in the rifampicin-based group, this would likely lead to an overestimation rather than an underestimation of success rates in the rifampicin group. The results of this study are in line with two

recent systematic reviews in which rifampicin-based strategies were not superior to non-rifampicin strategies[7,8]. The rationale behind the immediate start of the 5-day rifampicin treatment in our region is that the need for a highly bactericidal drug is expected to be most crucial in the early postoperative period after debridement. Rifampicin kills bacteria, including intracellular staphylococci, at a fast rate[19]. Experimental animal models showed that four days of rifampicin combination therapy quickly eradicated implant-associated infections[20]. The RCT in which treatment duration with rifampicin was 3-6 months was regarded as too heavily underpowered to implement long-term rifampicin treatment in our region. Therefore, a five day treatment schedule with rifampicin was chosen to quickly reduce the bacterial load around the implant in the early postoperative period. This should prevent new staphylococcal biofilm formation on the implant and so reduce the odds for a relapse. An important question that arises from our results is whether the first five days of rifampicin contributed at all to the high cure rates in the short-term rifampicin groups. This study cannot answer this question as patients were not treated *without* these five days of rifampicin. The attributive role of long-term rifampicin will be investigated in a large nationwide randomized controlled trial in The Netherlands.

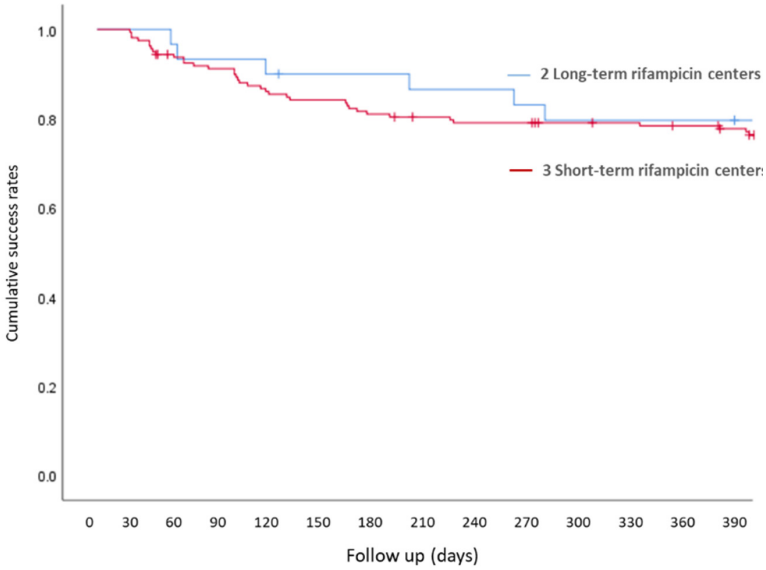


Figure 4. Comparison of success rates for all staphylococcal PJI stratified for centers with a long-term or a short-term rifampicin strategy*.

* Success rates were compared in 31 patients treated in a long-term rifampicin treatment center (in which protocol advised 12 weeks rifampicin combination therapy) and 169 patients treated in a short-term rifampicin center (in which protocol advised targeted monotherapy including only 5 days rifampicin combination therapy (with total treatment duration between six and twelve weeks).

Duration of antimicrobial therapy

Median duration of antibiotic therapy was four weeks shorter in the flucloxacillin- and clindamycin-based groups compared to the rifampicin-based group but with equal cure rates. Success rates were similar after splitting the flucloxacillin- and clindamycin-based groups in two groups based on treatment duration. Success rate was 82.6% if treated for 6 weeks (median treatment duration 40 days) and 86.3% if treated for >6 weeks (median treatment duration 63 days, p = 0.75). These results contradict the results of the recently published DATIPO trial in which twelve weeks of antimicrobial therapy was clearly superior to six weeks[21]. In the DATIPO trial, patients were randomized at the start of the study. In our cohort, the decision to quit antibiotics in the short-term group was made in the sixth week of treatment which has the advantage that the clinical course of the first six weeks could be considered (Table 1). Therefore, our data suggest, in line with other studies, that the decision to stop antimicrobial therapy after six weeks, based on a quickly improved clinical course, a normalized CRP and after MDT discussion, may still be regarded as a safe strategy[21-24].

Strengths and limitations

A major strength of this study is that several well-defined strategies were compared. Comparing one well-defined strategy (e.g., rifampicin, or clindamycin) with all other nón-defined strategies (e.g., non-rifampicin, or non-clindamycin) will usually lead to bias in favor of the well-defined strategy and may lead to unjustified rejection of equally good alternatives within that non-defined group (example of this is shown in Supplemental Figure 2). One possibility to solve this is to define several well-defined groups as was done in this study. However, confounding by indication can still be present in the well-defined groups as discussed for the clindamycin and flucloxacillin groups. Of note, this study also contains a fifth 'non-defined' group of patients, evenly present in all participating centers, with a worse outcome. Different treatment strategies within this group were very heterogeneous (Table 2).

To further strengthen the methodological quality of the study, patients with failure within two weeks after surgery (n=10, evenly distributed among the centers) were excluded from survival analysis. This is because these patients were still on intravenous antibiotics and had not yet started one of the preferred treatment options. Patients with megaprostheses (used in malignancies) were also excluded to reduce bias. Further, a second DAIR during treatment was not automatically considered a failure and resulted in cure in many patients (Table 3). If we would have defined all subsequent surgeries as failure, the overall cure rate would drop from 77% to 55% (Figure 2B). However, this drop in cure rate would evenly affect cure rates among all five antibiotic groups. These differences in cure show the importance of a uniform and clear-cut definition when comparing outcome between PJI studies. We suggest defining subsequent surgery only as a failure if a third debridement was needed or if surgery is needed after finishing antimicrobial therapy.

A limitation of the current study is the heterogeneity by including also patients with chronic PJI and patients with on-stage revision surgery. We thought it was justified to do so because a DAIR can still be a good treatment option in patients with longer duration of symptoms, as reported recently[25]. Although patients with one stage revision surgery were treated with the same short-term or long-term rifampicin strategy in the different centers, the surgical strategy differs from that of a DAIR. Therefore, we repeated the survival analysis, leaving out patients after one-stage exchange. This did not affect outcome (Supplemental Figure 3).

To exclude that the results of this study may be explained by other antibiotics that were used for pathogens in the patients with polymicrobial PJI, we performed an extra survival analysis including only the 130 patients with monobacterial staphylococcal PJI. This resulted in a limited increase in success rate in the vancomycin group (69% to 72%) and no change of success rates in the rifampicin-, flucloxacillin- and clindamycin-based groups, indicating that the activity against staphylococci was probably not caused by other antibiotics.

Summary and future perspectives

This study suggests that clindamycin or flucloxacillin monotherapy with only short-term induction therapy with rifampicin for five days might be considered as a reliable alternative to long-term rifampicin combination therapy. Although adjustment for confounding variables reduced bias as much as possible, the number of patients in the subgroups was still quite low. Future studies should assess whether adjunctive short-term induction therapy with rifampicin in patients treated with clindamycin or flucloxacillin has a significant impact on outcome. A large randomised controlled trial is warranted to definitively confirm the safety and effectivity of clindamycin and/or flucloxacillin monotherapy as appropriate alternatives for rifampicin combination therapy for staphylococcal PJI.

Acknowledgments

We are greatly indebted to Josefien Hessels and Maxime Gerritsen who had a central role in importing patient data into the Castor database. We also like to acknowledge the support from local research coordinators Menno Benard, Jantsje Pasma, Bregje Thomassen and Marjolein Schager with their kind help during the setup and follow up of the quality registry.

Funding

This work was supported by an unrestricted grant from the Dutch insurance company Zorg & Zekerheid [grant number ST.2015.53]

Conflicts of interests

The authors declare that they have no conflicts of interests.

References

1. Sabah SA, Alvand A, Price AJ. Revision knee replacement for prosthetic joint infection: Epidemiology, clinical outcomes and health-economic considerations. *Knee* 2021;28:417-21. doi: 10.1016/j.knee.2020.12.024 [published Online First: 2021/01/28]
2. RIVM. Report Postoperative Wound Infetions The Netherlands 2015-2019 2019 [Available from: https://www.rivm.nl/documenten/referentiecijfers-powi-2019.
3. Manning L, Metcalf S, Clark B, et al. Clinical Characteristics, Etiology, and Initial Management Strategy of Newly Diagnosed Periprosthetic Joint Infection: A Multicenter, Prospective Observational Cohort Study of 783 Patients. *Open Forum Infect Dis* 2020;7(5):ofaa068. doi: 10.1093/ofid/ofaa068 [published Online First: 2020/05/21]
4. Osmon DR, Berbari EF, Berendt AR, et al. Diagnosis and management of prosthetic joint infection: clinical practice guidelines by the Infectious Diseases Society of America. *Clin Infect Dis* 2013;56(1):e1-e25. doi: cis803 [pii];10.1093/cid/cis803 [doi]
5. Nguyen S, Robineau O, Titecat M, et al. Influence of daily dosage and frequency of administration of rifampicin-levofloxacin therapy on tolerance and effectiveness in 154 patients treated for prosthetic joint infections. *Eur J Clin Microbiol Infect Dis* 2015;34(8):1675-82. doi: 10.1007/s10096-015-2404-z [doi]
6. Vollmer NJ, Rivera CG, Stevens RW, et al. Safety and Tolerability of Fluoroquinolones in Patients with Staphylococcal Periprosthetic Joint Infections. *Clin Infect Dis* 2021;73(5):850-56. doi: 10.1093/cid/ciab145 [published Online First: 2021/02/20]
7. Scheper H, Gerritsen LM, Pijls BG, et al. Outcome of Debridement, Antibiotics, and Implant Retention for Staphylococcal Hip and Knee Prosthetic Joint Infections, Focused on Rifampicin Use: A Systematic Review and Meta-Analysis. *Open Forum Infect Dis* 2021;8(7):ofab298. doi: 10.1093/ofid/ofab298 [published Online First: 2021/07/15]
8. Aydin O, Ergen P, Ozturan B, et al. Rifampin-accompanied antibiotic regimens in the treatment of prosthetic joint infections: a frequentist and Bayesian meta-analysis of current evidence. *Eur J Clin Microbiol Infect Dis* 2021;40(4):665-71. doi: 10.1007/s10096-020-04083-4 [published Online First: 2020/10/31]
9. Theil C, Schmidt-Braekling T, Gosheger G, et al. Clinical use of linezolid in periprosthetic joint infections - a systematic review. *Journal of bone and joint infection* 2020;6(1):7-16. doi: 10.5194/jbji-6-7-2020 [published Online First: 2020/09/29]
10. Karlsen OE, Borgen P, Bragnes B, et al. Rifampin combination therapy in staphylococcal prosthetic joint infections: a randomized controlled trial. *J Orthop Surg Res* 2020;15(1):365. doi: 10.1186/s13018-020-01877-2 [published Online First: 2020/08/30]
11. Scheper H, van Hooven D, van de Sande M, et al. Outcome of acute staphylococcal prosthetic joint infection treated with debridement, implant retention and antimicrobial treatment with short duration of rifampicin. *The Journal of infection* 2018;76(5):498-500. doi: 10.1016/j.jinf.2018.01.009 [published Online First: 2018/02/08]
12. Dijkmans AC, Kweekel DM, Balmforth C, et al. The simplified oral flucloxacillin absorption test: an accurate method to identify patients with inadequate oral flucloxacillin absorption. *Neth J Med* 2019;77(7):255-60. [published Online First: 2019/10/05]

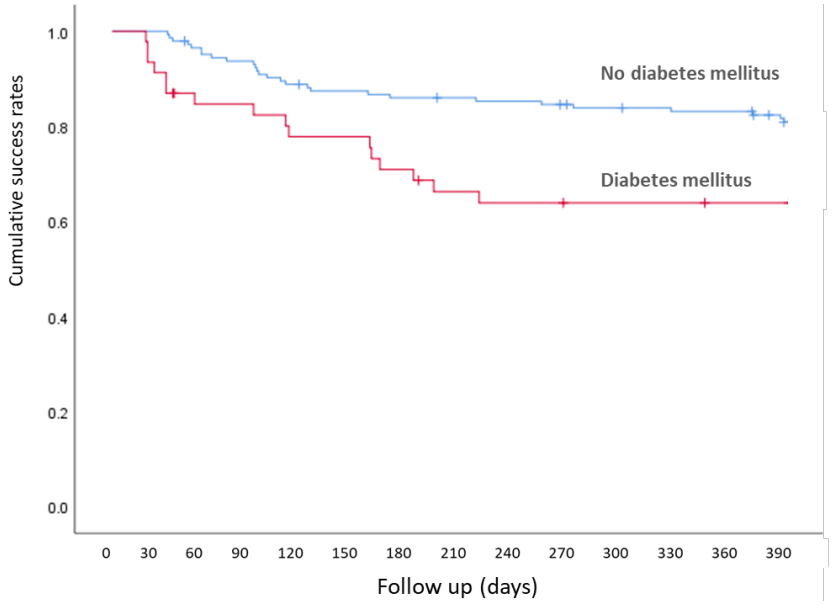
13. Tonnelier M, Bouras A, Joseph C, et al. Impact of rifampicin dose in bone and joint prosthetic device infections due to Staphylococcus spp: a retrospective single-center study in France. *BMC infectious diseases* 2021;21(1):174. doi: 10.1186/s12879-021-05832-2 [published Online First: 2021/02/14]
14. Thabit AK, Fatani DF, Bamakhrama MS, et al. Antibiotic penetration into bone and joints: An updated review. *International journal of infectious diseases : IJID : official publication of the International Society for Infectious Diseases* 2019;81:128-36. doi: 10.1016/j.ijid.2019.02.005 [published Online First: 2019/02/18]
15. Zeller V, Dzeing-Ella A, Kitzis MD, et al. Continuous clindamycin infusion, an innovative approach to treating bone and joint infections. *Antimicrob Agents Chemother* 2010;54(1):88-92. doi: 10.1128/AAC.01081-09 [published Online First: 2009/10/21]
16. Leijtens B, Elbers JBW, Sturm PD, et al. Clindamycin-rifampin combination therapy for staphylococcal periprosthetic joint infections: a retrospective observational study. *BMC infectious diseases* 2017;17(1):321. doi: 10.1186/s12879-017-2429-2 [published Online First: 2017/05/04]
17. Tevell S, Christensson B. Oral Flucloxacillin for Staphylococcal Osteomyelitis: Obsolete or Underused? *Journal of bone and joint infection* 2020;5(1):25-27. doi: 10.7150/jbji.42852 [published Online First: 2020/03/03]
18. Vilchez F, Martinez-Pastor JC, Garcia-Ramiro S, et al. Outcome and predictors of treatment failure in early post-surgical prosthetic joint infections due to Staphylococcus aureus treated with debridement. *Clin Microbiol Infect* 2011;17(3):439-44. doi: S1198-743X(14)63881-7 [pii];10.1111/j.1469-0691.2010.03244.x [doi]
19. Mandell GL, Vest TK. Killing of intraleukocytic Staphylococcus aureus by rifampin: in-vitro and in-vivo studies. *J Infect Dis* 1972;125(5):486-90. doi: 10.1093/infdis/125.5.486 [published Online First: 1972/05/01]
20. Zimmerli W, Sendi P. Role of Rifampin against Staphylococcal Biofilm Infections In Vitro, in Animal Models, and in Orthopedic-Device-Related Infections. *Antimicrob Agents Chemother* 2019;63(2) doi: 10.1128/AAC.01746-18 [published Online First: 2018/11/21]
21. Bernard L, Arvieux C, Brunschweiler B, et al. Antibiotic Therapy for 6 or 12 Weeks for Prosthetic Joint Infection. *N Engl J Med* 2021;384(21):1991-2001. doi: 10.1056/NEJMoa2020198 [published Online First: 2021/05/28]
22. Lora-Tamayo J, Euba G, Cobo J, et al. Short- versus long-duration levofloxacin plus rifampicin for acute staphylococcal prosthetic joint infection managed with implant retention: a randomised clinical trial. *International journal of antimicrobial agents* 2016;48(3):310-6. doi: 10.1016/j.ijantimicag.2016.05.021 [published Online First: 2016/08/16]
23. Chieffo G, Archambeau D, Eyrolle L, et al. Successful treatment of six weeks of antibiotics in hip and knee periprosthetic joint infection after one-stage replacement arthroplasty: a French cohort study. *Bone & Joint Journal Orthopaedic Proceedings Supplement* 2017;99-B(SUPP 22):88-88.
24. Bernard L, Legout L, Zurcher-Pfund L, et al. Six weeks of antibiotic treatment is sufficient following surgery for septic arthroplasty. *The Journal of infection* 2010;61(2):125-32. doi: 10.1016/j.jinf.2010.05.005 [published Online First: 2010/06/15]
25. Lowik CAM, Parvizi J, Jutte PC, et al. Debridement, antibiotics and implant retention is a viable treatment option for early periprosthetic joint infection presenting more than four weeks after index arthroplasty. *Clin Infect Dis* 2019 doi: 10.1093/cid/ciz867 [published Online First: 2019/09/11]

Supplemental files

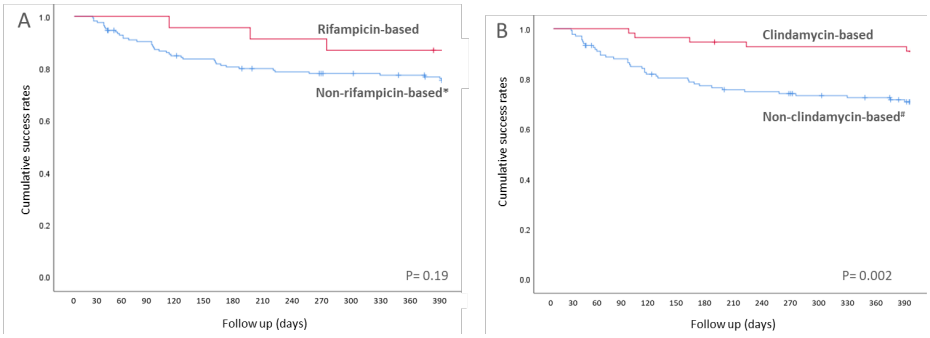
Supplemental table 1. Description of standardized protocol for Debridement procedure

Debridement of an acute prosthetic joint infection (PJI) should preferably start within 24 hours of (suspected) diagnosis. The operation proceeds according to the following step-by-step plan:

1. Preparation and protocol in theatre as for implantation of prosthetic joint
2. Antibiotic prophylaxis against postoperative wound infections is postponed until intraoperative cultures are taken.
3. Always perform an open arthrotomy. An arthroscopy in case of acute PJI is contraindicated.
4. Before starting debridement and antibiotics, 5-6 deep cultures are taken at the site of infection (fluid, tissue, capsule, synovia and bone, especially at the interphase). Culturing subcutaneous tissue cultures or wound smears are not indicated. Cultures are incubated for 14 days.
5. Cultures that are taken are placed directly into a sterile jar. Punctate fluids are directly inserted into a blood culture bottle. Change instrumentarium after each culture that is taken.
6. Cultures should be at the clinical microbiology laboratory as soon as possible.
7. After cultures are taken, extensive debridement takes place with excision of all "suspicious" or necrotic tissue and, if possible, a broad synovectomy. Exchangeable components of the prosthetic joint are removed, to allow for proper debridement of the joint, and replaced with new components.
    a. During exchange of mobile parts, the wound is first completely debrided and rinsed. Then, change gloves, disinfect skin with chlorhexidine, cover with clean covering material and clean instruments for insertion of prosthetic components, and close wound.
8. Next, rinse the prosthesis in situ with at least 6 L of Sodiumchloride 0.9% and use pulsavac. Use a wet gauze to "polish" the prosthetic parts in order to remove the formed glycocalyx macroscopically as much as possible. After 3-4 litres rinse with pulsavac and povidone iodine, then rinse the last litres with NaCl pulsavac.
9. Do not use gentamicin beads or gentamicin mats.
10. The wound is closed and no drains are left behind.



Supplemental Figure 1. Commparison of success rates for staphylococcal PJI between patients with and without diabetes mellitus.

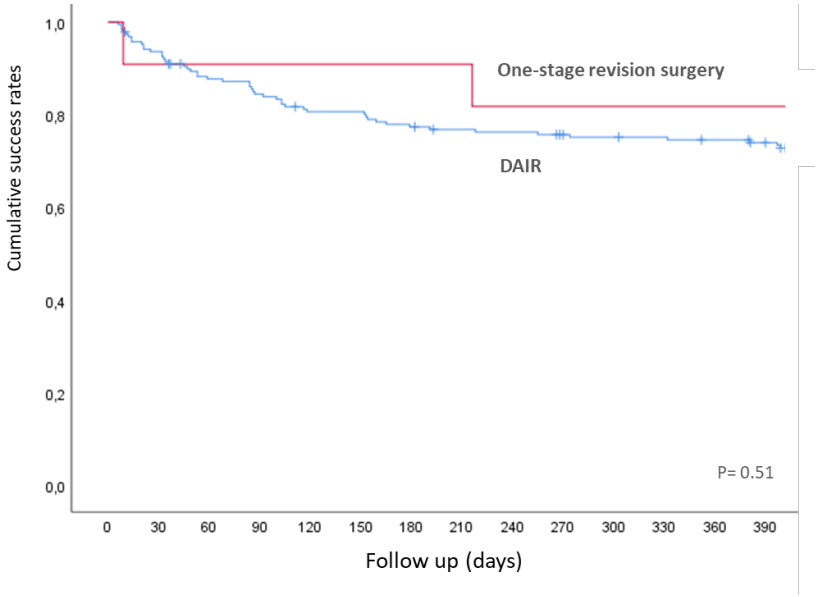


Supplemental Figure 2. Differences in success rates for staphylococcal PJI between well-defined and non-defined antibiotic groups.

This example shows how misinterpretation may occur if well-defined antimicrobial strategy is compared with a non-defined strategy. All failures within two weeks were excluded from this analysis. In graph A, the effectivity of rifampicin compared to non-rifampicin treatment is shown. However, the non-rifampicin group contains all patients treated with clindamycin-based strategy, but clindamycin is shown to be superior to non-clindamycin treatment in graph B. Stratification of the non-defined group may demonstrate potentially effective alternative treatment options.

*Non-rifampicin: all patients who were not treated in the rifampicin-based group.

Non-clindamycin: all patients who were not treated in the clindamycin-based group.



Supplemental Figure 3. Differences in success rates for patients treated with One-stage revision surgery (n=11) or DAIR (n=189) for staphylococcal PJI.

CHAPTER 6a

Outcome of acute staphylococcal prosthetic joint infection treated with debridement, implant retention and antimicrobial treatment with short duration of rifampicin

Henk Scheper, Daphne van Hooven, Michiel A.J. van de Sande, Robert J.P. van der Wal, Martha T. van der Beek, Leo G. Visser, Mark G.J. de Boer, Rob G.H.H. Nelissen

J Infect. 2018 May;76(5):498-500

Letter to the editor

A prosthetic joint infection (PJI) is a devastating complication of orthopaedic implant surgery. Cure with retention of an acutely infected prosthesis is possible if antimicrobial therapy is combined with thorough surgical debridement. Current international treatment guidelines for acute staphylococcal PJI advocate at least 12 weeks of combination therapy including rifampicin[1]. The evidence for shorter antimicrobial treatment duration with rifampicin is limited. Often, rifampicin is withheld until antimicrobial susceptibility is known and the postoperative wound is dry. Rifampicin might be most effective during the first days after debridement, the time period in which new biofilm formation on the surface of the implant needs to be prevented. Therefore, over the last 14 years, in our tertiary institution for orthopaedic implant surgery, all patients with an acute staphylococcal PJI who underwent a DAIR (Debridement, Antibiotics and Implant Retention) were treated with only five days of rifampicin in combination with at least 6 weeks of betalactam/glycopeptide antibiotics, both started immediately postoperative.

In this letter, we report the clinical outcome of these patients and assessed whether intraoperative start of rifampicin induced rifampicin resistance in patients who developed a relapse. Oncology patients with an infected megaprosthesis were also included. Patients were excluded if more than one prosthetic joint was infected. PJI was defined according to the IDSA criteria[1]. The criterion for 'acute' infection (three weeks) was extended to two months as DAIR was also performed in patients with longer duration of symptoms. The primary outcome was cure, defined as absence of infection and a stable retained implant for at least six months after stopping antibiotics. Failure was defined as either chronic suppressive antibiotic therapy with implant retention or removal of the implant. Treatment consisted of extensive surgical debridement, rinsing with povidone iodine and pulsed lavage with at least 3 liters of saline. Standard procedure required 3-6 periprosthetic tissue samples to be taken for culture. Empiric antibiotic therapy with a betalactam, an aminoglycoside and rifampicin (600 mg b.d.) was started intraoperative, after debridement. Rifampicin was stopped after five days. After two weeks, intravenous antibiotics were switched to an oral alternative depending on antimicrobial susceptibility testing, flucloxacillin oral absorption test[2] and the clinical response. Total treatment duration of six to twelve weeks depending on clinical response and inflammatory parameters. Follow up was at least one year.

Forty-one patients were included; baseline characteristics are shown in table 1. *Staphylococcus aureus* was involved in 30 cases and coagulase-negative staphylococci (CNS) in 10 cases. One patient had both a *S. aureus* and a CNS. In table 2 cure rates as categorized by affected joint, type of prosthesis and use of immunosuppression are summarized. Overall cure rate was 63%. Notably, patients without a megaprosthesis with a staphylococcal hip PJI (n=18) had a cure rate of 83%. Mean antimicrobial treatment duration in cured patients was 9.7 weeks (median 7.1 weeks). Twelve patients were treated for six weeks; their cure rate was 83%. Mean follow up of cured patients was 392 days (range 97-802 days).

Table 1. Baseline characteristics of 41 patients with acute staphylococcal PJI

	All (n=41)
Demographics	
Age at diagnosis (mean, range)	58 (15-92)
Sex (male, %)	24 (59%)
Implant site (n, %)	
Hip	22 (54%)
Knee	19 (46%)
Revision[#] (n, %)	14 (34%)
Comorbidities (n, %)	
Diabetes mellitus	4 (10%)
Rheumatoid arthritis	9 (22%)
Orthopaedic oncology[$]	14 (34%)
Use of immunosuppressant's^	11 (27%)
Clinical characteristics (n, %)	
Bacteraemia	9 (22%)
Duration of symptoms	
1-7 days	30
8-14 days	6
15-21 days	1
22-29 days	2
29-60 days	2
Microbiology	
Number of cultures taken (median, range)	5 (2-9)
Number of positive cultures per patient[@] (median, range)	4 (0-8)
Microbiology§	
S aureus	31 (76%)
CNS	10 (24%)

[#] patients with revision preceding PJI

^ Use of any of MTX/TNFa-inhibitors/steroids in the months preceding PJI

[$] patients with a tumour prosthesis in situ

[@] Two patients with evident pus but cultures remaining negative

Table 2. Subgroup analyses of outcome of DAIR and 5 days of rifampicin for acute staphylococcal PJI

	n	Complete cure[#]	Functional cure[*]
All patients	41	63%	76%
Patients without tumour prosthesis	27	70%	78%
Hip PJI	18	83%	89%
knee PJI	9	44%	56%
Patients with a megaprosthesis[#]	14	50%	71%
All patients with steroids/anti-TNF/MTX	11	46%	55%

^ acute: symptoms or last operation/revision < 8 weeks.

[#] Complete cure: absence of infection and a stable retained implant for at least six months after stopping antibiotic therapy

[*] Functional cure: stable prosthesis in situ but with chronic suppressive antimicrobial therapy

[#] Mega prosthesis: patients with bone- or soft-tissue tumors

Of the 15 failures, five had a functional (retention of the prosthesis with chronic suppressive therapy). Eight of those failures were caused by the same type of micro-organism as the primary infection (six *S aureus*, two CNS). Rifampicin susceptibility in seven of those latter cases had not changed. In the eighth patient one out of five positive cultures with *S aureus* showed rifampicin resistance.

The high cure rate for staphylococcal hip PJI exceeded those for knee PJI as observed in previous cohort studies.[3,4] Proportion of megaprostheses was higher in knee PJI (53%) compared to hip PJI (18%), which might explain differences in cure rate. The overall cure rate of 63% may be caused by group heterogeneity with respect to underlying disease (i.e. rheumatoid arthritis, bone- and soft-tissue tumors). Also, changing the liner or femoral head was not a routine procedure until three years ago, which might have decreased the likelihood for cure in our patients as well. Antibiotic treatment duration of six weeks was not associated with an increased relapse rate. Allegedly, clinicians are able to select patients who can be treated with a shorter course of antibiotic treatment, based on clinical and laboratory parameters.

The current advocated treatment policy for acute staphylococcal PJI is based on a small randomized trial in which patients were treated with rifampicin combination therapy for at least 12 weeks.[5] Of note, in this underpowered study 50% of the patients had osteosynthesis-associated infection and not a PJI. The drop-out rate due to rifampicin-related adverse events was 33% (6/18). Cure rates with combination therapy were thereafter reported to be 65-90% in observational studies[3-8]. No studies have been published in which short treatment duration with rifampicin was investigated. Our data suggest that prolonged treatment with rifampicin might not be needed as its added bactericidal and biofilm-preventing effect has already taken place in the first few postoperative days.[9] Rifampicin monotherapy and high bacterial loads are well known risk factors for evolving resistance. The absence of development of rifampicin resistance might be explained by the short treatment duration with rifampicin. However, resistance usually develops within two to three days of starting rifampicin monotherapy.[10]

This study adds new insights to the concept of antimicrobial treatment for patients with a staphylococcal PJI. Short-term postoperative treatment with rifampicin resulted in high cure rates in patients with staphylococcal hip PJI. Immediate intraoperative start of combination therapydid not result in rifampicin resistance. Additional prospective studies are warranted to elucidate the optimal duration of rifampicin as part of the antimicrobial therapy in patients with a staphylococcal PJI.

Funding: This research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors.

Conflicts of interest: none.

Reference list

1. Osmon DR, Berbari EF, Berendt AR, et al. Diagnosis and management of prosthetic joint infection: clinical practice guidelines by the Infectious Diseases Society of America. *Clin Infect Dis* 2013;56(1):e1-e25. doi: cis803 [pii];10.1093/cid/cis803 [doi]
2. Dijkmans AC, Hartigh J, van Dissel JT, et al. A simplified oral flucloxacillin absorption test for patients requiring long-term treatment. *Ther Drug Monit* 2012;34(3):356-58. doi: 10.1097/FTD.0b013e318257e8ac [doi]
3. Soriano A, Garcia S, Bori G, et al. Treatment of acute post-surgical infection of joint arthroplasty. *Clin Microbiol Infect* 2006;12(9):930-33. doi: S1198-743X(14)64316-0 [pii];10.1111/j.1469-0691.2006.01463.x [doi]
4. Barberan J, Aguilar L, Carroquino G, et al. Conservative treatment of staphylococcal prosthetic joint infections in elderly patients. *Am J Med* 2006;119(11):993-10. doi: S0002-9343(06)00572-9 [pii];10.1016/j.amjmed.2006.03.036 [doi]
5. Zimmerli W, Widmer AF, Blatter M, et al. Role of rifampin for treatment of orthopedic implant-related staphylococcal infections: a randomized controlled trial. Foreign-Body Infection (FBI) Study Group. *JAMA* 1998;279(19):1537-41. doi: joc71805 [pii]
6. Lora-Tamayo J, Murillo O, Iribarren JA, et al. A large multicenter study of methicillin-susceptible and methicillin-resistant Staphylococcus aureus prosthetic joint infections managed with implant retention. *Clin Infect Dis* 2013;56(2):182-94. doi: cis746 [pii];10.1093/cid/cis746 [doi]
7. Vilchez F, Martinez-Pastor JC, Garcia-Ramiro S, et al. Outcome and predictors of treatment failure in early post-surgical prosthetic joint infections due to Staphylococcus aureus treated with debridement. *Clin Microbiol Infect* 2011;17(3):439-44. doi: S1198-743X(14)63881-7 [pii];10.1111/j.1469-0691.2010.03244.x [doi]
8. Ascione T, Pagliano P, Mariconda M, et al. Factors related to outcome of early and delayed prosthetic joint infections. *J Infect* 2015;70(1):30-36. doi: S0163-4453(14)00204-7 [pii];10.1016/j.jinf.2014.07.008 [doi]
9. Zimmerli W, Frei R, Widmer AF, et al. Microbiological tests to predict treatment outcome in experimental device-related infections due to Staphylococcus aureus. *J Antimicrob Chemother* 1994;33(5):959-67.
10. Kadurugamuwa JL, Sin LV, Yu J, et al. Noninvasive optical imaging method to evaluate postantibiotic effects on biofilm infection in vivo. *Antimicrob Agents Chemother* 2004;48(6):2283-87. doi: 10.1128/AAC.48.6.2283-2287.2004 [doi];48/6/2283 [pii]

CHAPTER 7

SAAP-148 eradicates MRSA persisters within mature biofilm models simulating prosthetic joint infection

Henk Scheper, Julia M. Wubbolts, Joanne A.M. Verhagen, Adriëtte W. de Visser, Robert J.P. van der Wal, Leo G. Visser, Mark G.J. de Boer, Peter H. Nibbering

Frontiers in Microbiology 2021 Jan 29;12:625952

Abstract

Prosthetic joint infection (PJI) is a severe complication of arthroplasty. Due to biofilm and persister formation current treatment strategies often fail. Therefore, innovative anti-biofilm and anti-persister agents are urgently needed. Antimicrobial peptides with their broad antibacterial activities may be such candidates. An *in vitro* model simulating PJI comprising of rifampicin/ciprofloxacin-exposed, mature methicillin-resistant *Staphylococcus aureus* (MRSA) biofilms on polystyrene plates, titanium/aluminium/niobium discs and prosthetic joint liners was developed. Bacteria obtained from and residing within these biofilms were exposed to SAAP-148, acyldepsipeptide-4, LL-37 and pexiganan. Microcalorimetry was used to monitor the heat flow by the bacteria in these models. Daily exposure of mature biofilms to rifampicin/ciprofloxacin for 3 days resulted in a 4-log reduction of MRSA. Prolonged antibiotic exposure did not further reduce bacterial counts. Microcalorimetry confirmed the low metabolic activity of these persisters. SAAP-148 and pexiganan, but not LL-37, eliminated the persisters while ADEP4 reduced the number of persisters. SAAP-148 further eradicated persisters within antibiotics-exposed, mature biofilms on the various surfaces. To conclude, antibiotic-exposed, mature MRSA biofilms on various surfaces have been developed as *in vitro* models for PJI. SAAP-148 is highly effective against persisters obtained from the biofilms as well as within these models. Antibiotics-exposed, mature biofilms on relevant surfaces can be instrumental in the search for novel treatment strategies to combat biofilm-associated infections.

Introduction

Yearly over one million prosthetic joints are implanted in patients in the United States. Prosthetic joint infection (PJI) is a severe complication occurring in 1-3% of patients and has a high economic burden on health care systems. Most PJIs are caused by staphylococci[1] [2]. Treatment of patients with an acute PJI consists of thorough surgical debridement of the implant and the infected tissue around it, followed by 6-12 weeks of antibiotic therapy. Nevertheless, failure rates for this treatment strategy are considerable, ranging from 10% to 45% in some of the largest studies[3] [4]. An important cause of treatment failure is the formation of a biofilm on the surface of the implant. A biofilm is formed by bacteria that, after adherence to the implant, form a matrix of extrapolymeric substances (EPS) that protect bacteria against the actions of antibiotics and effectors of host's immune systems[5]. Within biofilms bacteria may switch phenotypically to a metabolically inactive, non-dividing, dormant state, called persisters[5] [6]. Persisters are defined as metabolically inactive, dormant bacteria that survive lethal concentrations of antibiotics without induction of resistance. The formation of persisters is triggered by stress factors like lack of nutrients and exposure to antibiotics [7]. Persisters within biofilms are tolerant to antibiotic therapy, which contributes to PJI treatment failures [8]. Based on these considerations, innovative anti-biofilm and anti-persister treatment strategies are urgently needed. For evaluation of such candidates an *in vitro* biofilm model that approximates a PJI as closely as possible is instrumental.

Antimicrobial peptides are considered promising candidates to combat biofilm-associated infections. For instance, the human cathelicidin LL-37 has broad-spectrum antibacterial activities, including antibiofilm activity, together with immune modulating capabilities[9]. SAAP-148, a synthetic peptide based on LL-37, has shown to be more effective in eradicating bacteria than LL-37[10]. Acyldepsipeptide 4 (ADEP4) activates bacterial proteases in an ATP-independent manner resulting in cell death. In combination with rifampicin, ADEP4 eradicates biofilms in a mouse model with a chronic *S. aureus* infection[8]. Pexiganan, an analogue of magainin isolated from the skin of the African clawed frog, exhibited *in vitro* broad-spectrum antibacterial activity[11].

For the current study we developed an *in vitro* biofilm model approximating a prosthetic joint infection as closely as possible. With this model the efficacy of four promising antimicrobial peptides on persisters and bacteria in other growth modes in mature biofilms was assessed.

Materials and Methods

Antibiotics and antimicrobial peptides

Ciprofloxacin hydrochloride (Sigma-Aldrich, PHR 1044-1G) and rifampicin (Sigma-Aldrich, R3501-250 mg) at concentrations corresponding to 10x the minimal bactericidal concentration (MBC) for MRSA LUH14616 were used (1.28 mg/mL ciprofloxacin, 10 µg/mL rifampicin). The MBC was defined as the lowest concentration that killed 99.9% of the bacteria compared to untreated control bacteria. SAAP-148 (LKRVWKRVFKLLKRYWRQLKKPVR), LL-37 (LLGDFFRKSKEKIGKEFKRIVQRIKDFLRNLVPRTES), and pexiganan (GIGKFLKKAKKFGKAFVKILKK), all N-terminal acetylated, C-terminal amidated, were synthesized by solid phase strategies on an automated multiple peptide synthesizer (SyroII, MultiSyntech, Witten, Germany) as described elsewhere [10 12]. The molecular mass of the peptides was confirmed by mass spectrometry and the purity of the peptide exceeded 95%, as determined by reverse phase high-performance liquid chromatography. The lyophilized peptides were stored at -20°C until use. The Clp protease activator acyldepsipeptide 4 (ADEP4) was purchased from ABGENT, a WuXi AppTec company (China); the purity of this commercial peptide was >98%. Peptide stocks were stored in 0.01% acetic acid (pexiganan), dimethyl sulphoxide (ADEP4) and milliQ (SAAP-148 and LL-37). For experiments, peptide stocks were diluted in phosphate-buffered saline (PBS) to the desired concentrations.

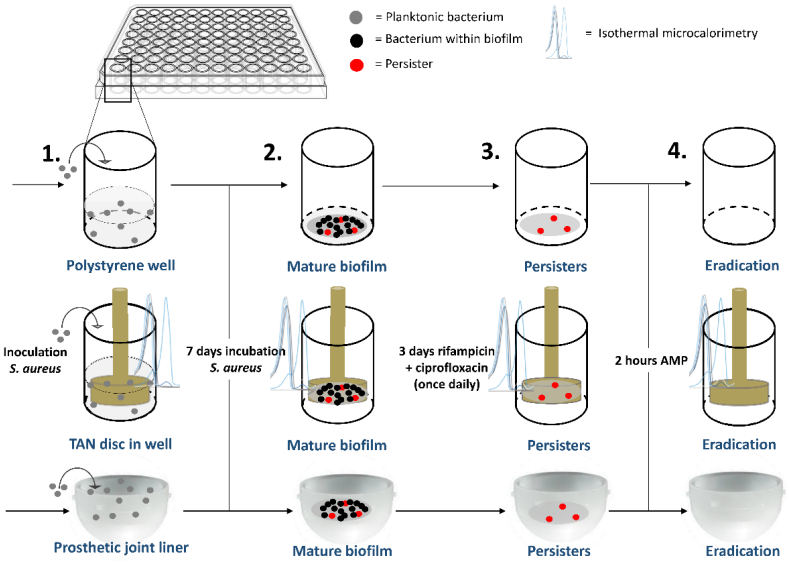
Methicillin-resistant *Staphylococcus aureus* (MRSA)

MRSA LUH14616 sequence type 247, was collected by a nasal swab from a patient without an infection. It was preserved in nutrient broth supplemented with 20% glycerol at -80˚C. Prior to experiments, inocula from the frozen stocks were grown overnight at 37˚C on sheep blood agar plates (BioMerieux). Thereafter, bacteria were cultured to mid-log phase in tryptic soy broth for 2.5 h at 37˚C. Finally, the bacteria were harvested by centrifugation (1,000 x g for 10 min) and then resuspended in PBS to the desired inoculum concentration (10^7 CFU/mL), based on the optical density at 600 nm.

In vitro biofilm model simulating biofilm associated infection

Mid-log phase MRSA were diluted to 10^7 CFU/mL in brain heart infusion (BHI) medium. Next, 100 µl of this suspension were cultured in 96-wells polystyrene plates covered with breathable seals and incubated at 37˚C for 7 days in a humidified environment. Thereafter, the medium was removed and the wells were washed twice with phosphate-buffered saline (PBS, 140 mM NaCl, pH 7.4). Next, 100 µL of fresh BHI medium containing ciprofloxacin and rifampicin, both 10x MBC, was carefully added to each well in order not to disrupt the biofilm. The medium containing antibiotics was refreshed daily for 72 h. In the second model TAN discs (consisting of titanium7%-aluminium6%-niobium; ISO5832/11) were inserted in the 96-well plates and mature biofilms were developed on these metal discs using the protocol as above. A third model comprised of bacterial

biofilms developed on the bottom of the cup of a prosthetic hip liner. Twelve ultra-high-molecular-weight polyethylene acetabular cups were provided by Waldemar Link GmbH & Co. KG (Germany). During formation of the biofilm and antibiotic exposure, the liner was covered with aluminium foil to prevent contamination and dehydration of the biofilm. Liners were re-used due to the limited number of liners provided. Prior to re-use, the liner was, after rinsing with 70% ethanol, submerged in 70% ethanol overnight after which the liner was rinsed again with 70% ethanol and autoclaved thereafter. The experiments with the liners were done twice. In the first dose-finding experiment liners were tested per increasing SAAP-148 concentration. Thereafter, the experiment was repeated for the concentrations with the highest effectivity in the first experiment. A schematic overview of the different models is provided in Figure 1.



	Inoculation device	Biofilm formation	Antibiotic treatment	Anti-persister treatment
	Phase 1	Phase 2	Phase 3	Phase 4
Biofilm	Absent	Mature	Treated	Treated
Exposure	No	No	Antibiotics	AMP
Planktonic bacteria	++++	Few	*Dependent on effect*	None
Persisters	Very few	Few	Few	*Dependent on effect*
Viable cells	++	++	+	*Dependent on effect*

Figure 1. Overview of the mature, antibiotics-exposed biofilm model.
Briefly, methicillin-resistant *Staphylococcus aureus* (MRSA) were inoculated into a well, on a metal implant device within a well and into a sterile acetabular hip liner, incubated for seven days, exposed to rifampicin and ciprofloxacin and subsequently exposed to antimicrobial peptide. AMP: antimicrobial peptide. Estimated proportion of persisters/viable bacteria: +: <0.1%, ++: >0.1% and <1%, ++++: 10-100%

Assessment of the effects of antimicrobial peptides on persisters in antibiotics-exposed mature biofilms

The activity of antimicrobial peptides against bacteria, i.e. persisters and possibly other bacterial subpopulations with a long lag time, in antibiotics-exposed mature biofilms was assessed by exposure of the biofilms to increasing concentrations of antimicrobial peptides in PBS for 2 or 24 h after removal of the supernatant. Prior to and at the indicate interval after exposure to the peptide, biofilms were sonicated in PBS and the number of surviving bacteria was determined microbiologically. In case of complete eradication bacterial plates were inspected again after 5 days in the incubator for possible regrowth of persisters and/or bacteria with a long lag time. To assess the direct effects of the peptides on these bacterial subpopulations, antibiotics-exposed mature biofilms from multiple wells were sonicated, pooled and diluted in PBS and then exposed to increasing concentrations of antimicrobial peptides. The possibility that not all bacteria could be obtained from biofilms by sonication was investigated microbiologically and we did not find viable bacteria (even up to five days of maintaining the bacterial plates in the incubator) remaining in the wells**.** In addition, the viability of bacteria was also not affected by the sonication procedure used in these experiments.

Isothermal microcalorimetric (IMC) assay

Isothermal microcalorimetry was used to monitor heat flow (µW) by MRSA in four different stages during the formation of antibiotics-exposed mature biofilms on TAN discs in real time for 30 h using a Calscreener (Symcel Sverige, Spånga, Sweden). BHI broth was used as a reference to calibrate the Calscreener. Mature biofilms and antibiotics-exposed mature biofilms were developed in this model as described above. After removal of the supernatant by two washes with PBS the inserts containing the biofilms were transferred to metal microcontainers and then exposed to 100 µL of BHI with or without 51.2 µM SAAP-148 peptide. In addition, 100 µL of $1x10^7$ CFU planktonic MRSA/mL were transferred to metal microcontainers Furthermore, microcontainers with 100 µL of BHI served as a control. All metal microcontainers were maintained in the Calscreener for 4 days at 37 °C for continuous monitoring of the heat production by the various bacterial populations. At the end of the experiments the microcontainers were sonicated for 10 min and the number of persisters was determined microbiologically and cultured for five days afterwards.

Statistical analysis

TThe non-parametric Mann–Whitney U test was performed to determine statistical significance when comparing medians of antibiotics and/or antimicrobial peptide treated biofilms using GraphPad prism (GraphPad software, La Jolla, CA United States). P values ≤0.05 were considered to be statistically significant different.

Results

Effect of antibiotics on bacteria within mature biofilms on polystyrene

A constant bacterial load of 8log CFU/ml was present during the seven days of biofilm maturition (Figure 2A). Results revealed a time-dependent reduction in bacterial counts in seven-day mature antibiotics-exposed MRSA biofilms on polystyrene plates with a maximum 4 log reduction at day three-four (Figure 2B), indicating that the surviving bacteria were antibiotic-tolerant. Therefore, antibiotic exposure of mature biofilms for more than three days was considered sufficient for persister enrichment.

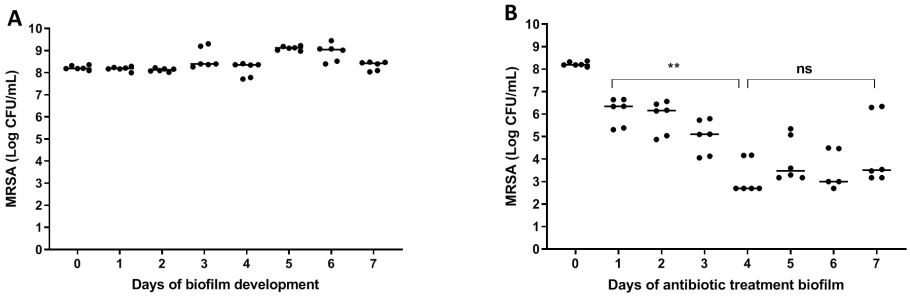


Figure 2. Effect of antibiotics on bacteria within mature biofilms on polystyrene.

A. The number of bacteria within biofilms on 96-well polystyrene plates remained constant for 7 days. **B**. Exposure of a seven-day mature biofilm on 96-well polystyrene plates to rifampicin/ciprofloxacin daily for three days, significantly reduced the bacterial load (p = 0.002). Prolonged exposure to rifampicin/ciprofloxacin did not result in further reduction of bacterial counts. The solid line denotes the median log CFU/mL. n=3 experiments, each in duplicate. NS = not significant.

MRSA in antibiotics-exposed mature biofilms are dormant

To further characterize these antibiotic-tolerant bacteria their metabolic activity was measured by isothermal calorimetry. Results revealed that heat flow of these bacteria in antibiotics-exposed, mature biofilms was almost zero (Fig. 3A-C), indicating that these bacteria are metabolically inactive, i.e. persisters. Of note, an initial modest peak of 7 µW upon incubation of these cells with BHI confirms their ability to revive. For comparison, we also assessed heat flow by planktonic MRSA and bacteria in mature biofilms. Results revealed two peaks in the heat flow curve of planktonic bacteria: the first peak occurred after 2.5 h of incubation and the second peak at 6-8 h (Fig. 3A). Thereafter, heat flow dropped to a value of approximately 10 µW, probably due to the evolution to stationary phase bacteria with less metabolic activity. MRSA within mature biofilms supplemented with BHI (bacterial load >5x10^8 MRSA) showed a peak in heat production around 6 h after which the level decreased to a continuous level of 10 µW, which was equal to heat production by stationary phase bacteria (Fig. 3B), indicating that heat production by bacteria within a biofilm is considerably less than by mid log phase bacteria.

Effect of SAAP-148, ADEP4, LL-37 and pexiganan on MRSA persisters

To select the most promising antimicrobial peptide, the direct effect of SAAP-148, ADEP4, LL-37 and pexiganan on persisters obtained from antibiotics-exposed, mature MRSA biofilms on polystyrene plates was assessed (Figure 4). Within 2 h SAAP-148 (at doses ≥ 1.6 µM; Figure 4A) and pexiganan (at doses ≥ 12.8 µM; Figure 4B) eradicated all bacteria, whereas bacterial counts were reduced by LL-37 (Figure 4C) and ADEP4 (Figure 4D). In agreement with the expectation that ADEP4 requires more time to exert its effects, we found that at 24 h of exposure all persisters were eliminated by ADEP4 (at doses ≥ 12.8 µM; Figure 4E). Of note, bacterial samples obtained after exposure to the peptides were cultured up to 5 days to ascertain that all persisters were killed. In addition, SAAP-148 was also highly effective in eliminating persisters residing in antibiotics-exposed, mature biofilms with complete eradication seen at ≥ 1.6 µM (Figure 3F). Based on these data SAAP-148 was selected for further experiments.

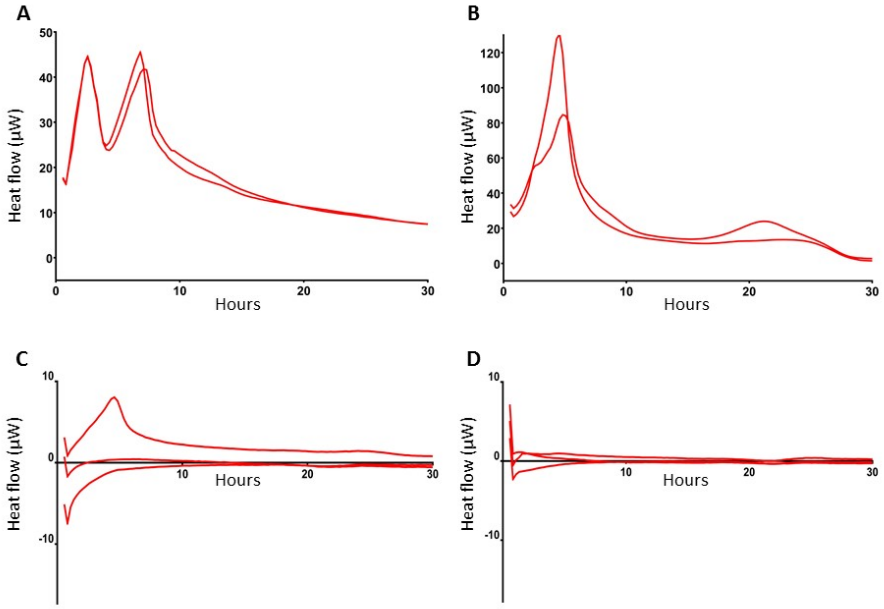


Figure 3. Heat flow by MRSA in log phase, in mature biofilms and in mature, antibiotics-exposed biofilms with and without exposure to SAAP-148.

Heat flow by $1x10^6$ log phase MRSA (A), $>5x10^8$ MRSA in a mature biofilm (B), $1x10^4$ persisters in antibiotics-exposed biofilms upon exposure to BHI broth (C) and antibiotics-exposed mature biofilms to BHI supplemented with 51.2 µM SAAP-148 (D). Log-phase MRSA, MRSA in mature biofilms and antibiotics-exposed biofilms were maintained in microcontainers in a Calscreener during 30 h. There was no heat flow in the persister subpopulation apart from a small peak after addition of BHI (C). Persisters exposed to SAAP-148 did not display any detectable heat flow (D). Results are from two replicates of a representative experiment (n=2-3 experiments).

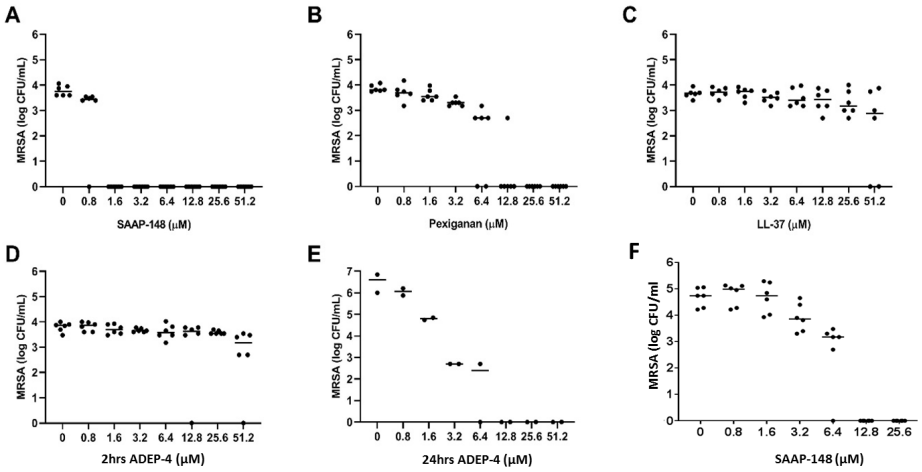


Figure 4. Effect of SAAP-148, ADEP4, LL-37 and pexiganan on MRSA originating from antibiotics-exposed seven-day mature biofilms.

SAAP-148 resulted in complete eradication of bacteria sonicated from antibiotics-exposed seven-day mature biofilms on polystyrene at doses ≥ 1.6 μM (**A**). Exposure of the intact biofilm without sonication to SAAP-148 resulted in complete eradication at doses ≥ 12.8 μM (**B**). Pexiganan resulted in complete eradication of sonicated bacteria at doses ≥ 6.4 μM (**C**). Acyldepsipeptide 4 (ADEP4) (**D**) reduced the bacterial counts by 1 log. Exposing the bacteria for 24 h to ADEP4 (**E**) resulted in compete eradication at doses ≥ 12.8 μM. Human cathelicidin LL-37 (**F**) reduced the bacterial counts by 1 log. Solid lines denote the median log CFU/mL. Fig. 3A-E: n=3 experiments (each in duplicate) Fig. 3F: n=1 experiment in duplicate. ns = not significant.

Effect of SAAP-148 on MRSA persisters obtained from and residing in antibiotics-exposed mature biofilms on TAN discs

Next, seven-days mature MRSA biofilms were produced on TAN discs and then exposed for three days to rifampicin and ciprofloxacin. SAAP-148 eliminated all persisters obtained from these antibiotics-exposed mature MRSA biofilms in a dose-dependent fashion with complete eradication already seen at ≥ 1.6 μM (Figure 5A). Exposure of the persisters residing in antibiotics-exposed mature biofilms to SAAP-148 also resulted in complete eradication, but at higher doses (≥ 51.2 μM; Figure 5B). Prolonged exposure of the persisters in biofilms to SAAP-148 did not improve the efficacy of the peptide (eradication at doses ≥ 51.2 μM; Figure 5C). To rule out the possibility that SAAP-148 was in fact effective against persisters which became metabolically active again after quitting antibiotic therapy, the experiment was repeated with addition of the antibiotics together with SAAP-148 on the fourth day of antibiotic exposure. This also resulted (in five out of six experiments) in elimination of all biofilm-embedded bacteria from a dose of 51.2 μM (Figure 5D). In agreement, calorimetry showed that SAAP-148 reduced heat production of bacteria residing in the biofilm on TAN discs to undetectable levels (Figure 3D). Together, these data indicate that higher doses of SAAP-148 are required to eliminate bacteria within the mature biofilm than when directly in contact with the persisters.

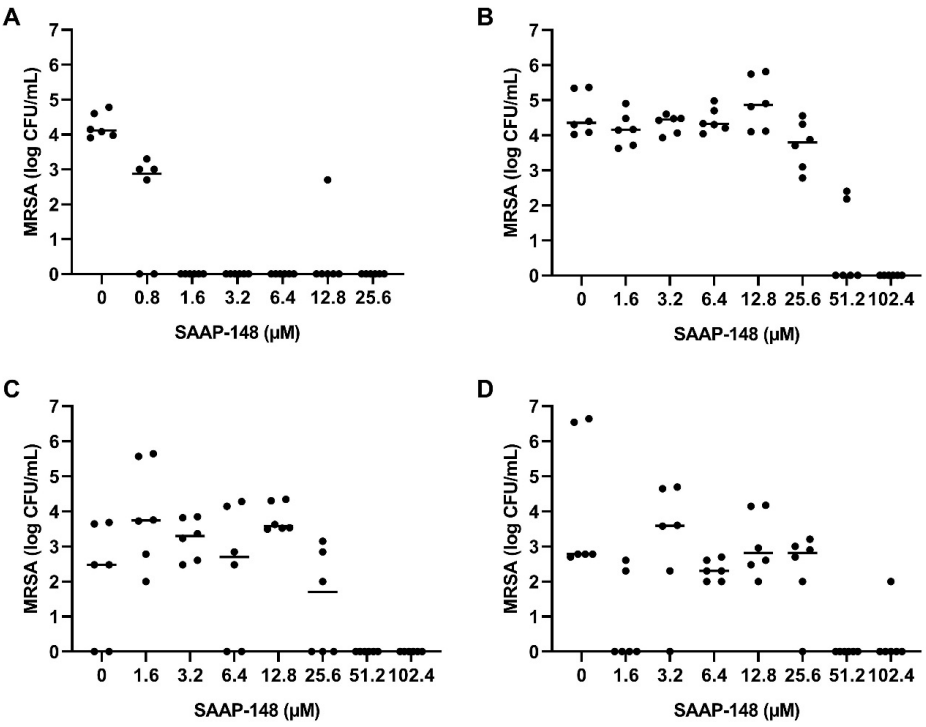


Figure 5. Effect of SAAP-148 on MRSA persisters in antibiotics-exposed mature biofilms on TAN discs.
Bacteria obtained by sonication from antibiotics-exposed mature biofilms on TAN discs were exposed for 2 h to SAAP-148. This resulted in eradiation of the bacteria at SAAP-148 doses of ≥1.6 μM (**A**). SAAP-148 dose-dependently reduced bacterial counts in intact biofilms (without sonication) on TAN discs at doses ≥ 51.2 μM (**B**). Prolonging the exposure of the biofilms to SAAP 148 to 24 h resulted in similar eradication of bacteria at doses ≥ 51.2 μM (**C**). Addition of SAAP 148 for 24 h during an additional fourth day of antibiotic exposure on the intact biofilm also resulted in eradication of bacteria (**D**). Solid lines denote the median log CFU/mL. (n=3 experiments, each in duplicate).

Effect of SAAP-148 on MRSA biofilms formed on a polyethylene insert of a prosthetic hip joint

To simulate a PJI more closely, the effect of different SAAP-148 concentrations was assessed on persisters in antibiotics-exposed, mature biofilms on sterile acetabulum liners of a hip prosthesis. Results revealed eradication of the bacteria by peptide at all concentrations ≥25.6 μM except for 3 outliers (Figure 6). The results indicate that SAAP-148 is also effective against persisters in mature biofilms on acetabular hip liners.

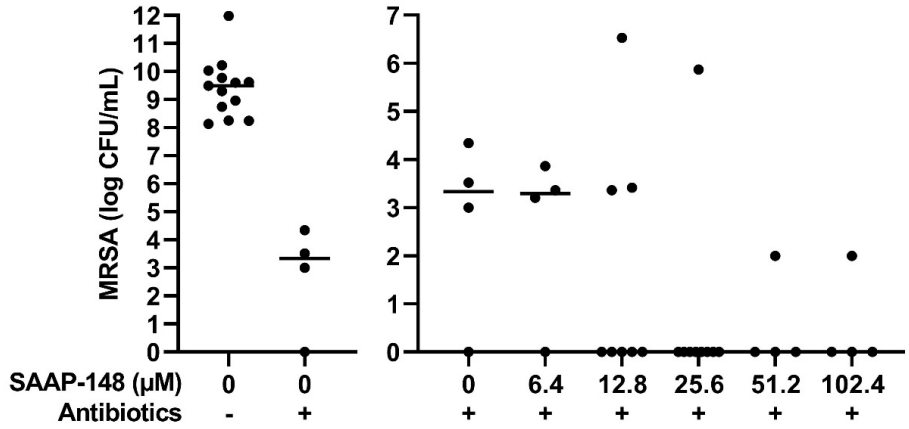


Figure 6. **Effect of SAAP-148 on mature antibiotics-exposed MRSA biofilms on an acetabular hip liner**. From a SAAP-148 dose of 25.6 µM, bacteria were eradicated from the majority of liners. This experiment was performed twice. In the second experiment, more liners were used for the concentrations with the highest effectivity in the first experiment. Controls and 6.4uM: n=4 liners; 12.8uM: n= 8 liners; 25.6 uM: n=9 liners; 51.2 and 102.4uM: n=4 liners. Outliers were present in both experiments. Antibiotics -: before exposure to antibiotics. Antibiotics +: after exposure to antibiotics. Solid lines denote the median log CFU/mL.

Discussion

Importance of an in vitro mature biofilm model simulating PJI and other foreign material infections

As antibiotic treatment of PJI often fails novel agents that eradicate persisters in mature biofilms are urgently needed. In this study we compared the anti-persister activities of synthetic antimicrobial peptides as potential candidates. For this purpose, we first developed innovative *in vitro* models simulating a PJI. These models were based on the following considerations. First, in most clinical biofilm-associated infections like PJI a mature biofilm has developed. The heterogeneous biofilm structure with extracellular polymers substances, eDNA and proteins makes drug penetration more difficult [13]. Also, the subpopulation of persisters in mature biofilms is denser than in immature biofilms (that are often used in biofilm studies) due to antibiotic treatment and nutrient starvation. To avoid outcomes that may not be optimal for translation to clinical biofilm-associated infections, we used seven-day matured MRSA biofilms followed by three days of antibiotic exposure. Second, we developed the biofilms on acetabular hip liners and metal alloys (titanium, niobium and aluminium) that are used in prosthetic joints . Third, to avoid awakening of persisters, exposure of the persisters to antimicrobial peptides is initiated immediately after termination of antibiotic exposure. Finally, we ruled out late regrowth of surviving persisters in biofilms after SAAP-148 exposure by inspecting the bacterial agar plates after 5 days in the incubator. These *in vitro* models can be used to screen other anti-biofilm and

anti-persister agents, although limitations in simulating PJI should be taken into account, such as the absence of host cells and inflammatory mediators.

Antibiotic tolerance of persisters to rifampicin and ciprofloxacin

We exposed mature biofilms to high doses of rifampicin and ciprofloxacin. These antibiotics are widely used as treatment for staphylococcal PJI, penetrate well in biofilms, and reduce bacterial counts within biofilms significantly [14]. We found that the antibiotics reduced the bacterial load in mature biofilms by >99.9% with the remaining bacteria displaying tolerance for high doses of rifampicin and ciprofloxacin. Microcalorimetry confirmed the dormant state of these antibiotics-tolerant bacterial cells as well as their ability to revive upon addition of bacterial growth medium. A limitation of microcalorimetry is its lower limit of detection being approximately 1×10^4 bacteria [15], which is close to the number of persisters in the mature biofilms. Also, the measured heat flow is the sum of all chemical and physical processes that take place within the bacterial community. Obviously, additional and more sensitive methods, for example transcriptome analysis, cryo-electron microscopy and/or measurement of ATP levels in bacteria within biofilms, should be implicated to further characterize the persisters. Nevertheless, we can conclude that substantial numbers of persisters are present in the current antibiotics-exposed, mature biofilms. We cannot exclude that bacteria with a long lag time or small colony variants (SCVs) also survived antibiotic exposure. SCVs differ from the normal phenotype in their small colony size and reduced growth rate. However, complete elimination of all bacterial cells that were not affected by the antibiotics indicates that these SCVs, if present, were also killed by the antimicrobial peptides [16 17]. The inability of rifampicin and ciprofloxacin to eradicate persisters is in line with other studies that showed incomplete eradication [8 18-21] or even induction of persisters [22]. Interestingly, rifampicin in combination with a fluoroquinolone eliminated all bacteria in several experimental animal models with foreign-body infections [14 23-25]. The strong innate immune response in these animals may have contributed to this favorable outcome. The favorable outcome may also be related to the maturation state of the biofilms as the rifampicin combination was not effective in 2-week MRSA biofilms in a rat model [26]. In a guinea pig tissue-cage infection model rifampicin eradicated implant-adhering *S. aureus* after a 12 h treatment delay but not after a 24-48 h treatment delay. [27]. Together, results from studies on elimination of bacteria in immature biofilms in animals with a strong innate immune response may not be representative for mature biofilms in human biofilm-associated infections.

Effectivity of antimicrobial peptides

The main conclusion from this study pertains to the efficacy of four promising antimicrobial peptides to eradicate persisters within antibiotics-exposed, mature biofilms. Both SAAP-148 and pexiganan rapidly eliminated biofilm-derived bacteria in a

dose-dependent fashion with SAAP-148 being the most effective peptide. The required concentration of SAAP-148 to eliminate persisters within biofilms was considerably higher than for direct killing of the persisters, indicating that peptide's antibacterial and antipersister activities are hampered by the extracellular matrix of the biofilm. Interestingly, higher SAAP-148 concentrations were needed for biofilms on TAN discs and hip liners, indicating that the surface of the implant may play a role in the development of the biofilm that protects bacteria within it. Despite displaying good antibiofilm activity in earlier studies, LL-37 reduced the bacterial counts in the antibiotics-exposed, mature biofilms only moderately. ADEP4 eliminated the bacteria in the biofilms in a dose-dependent fashion at 24 h, but not at 2 h of exposure. This was expected as it takes more time before bacteria die from massive protein breakdown due to activation of the ATP-independent caseinolytic protease Clp, the proteolytic core of a major bacterial protein degradation machinery, by ADEP [28-30]. Together, SAAP-148, ADEP4, LL-37 and pexiganan all exerted activity against MRSA persisters obtained from mature antibiotics-exposed, mature biofilms as well as persisters in such biofilms. The most effective peptide, SAAP-148, eliminated all persisters within mature biofilms on polystyrene, TAN discs and on most prosthetic hip liners. Unexpected survival of bacteria was seen in the experiments with the TAN discs and in both liner experiments (figure 5 and 6). Given the high bacterial load we presumed that these bacteria had not been exposed to the antimicrobial peptide in these experiments. Therefore, we regarded those as outliers related to outgrowth of untreated persisters. Of note, SAAP-148 killed the persisters as well as log phase bacteria within 2 h, indicating that peptide's toxic effect on bacteria is independent of the metabolic activity of bacteria[10]. SAAP-148 kills bacteria by binding to the phospholipid bilayer of the bacterial membrane and the subsequent conformational change of the peptide that causes direct leakage of bacteria resulting in cell death.

Antibiofilm effects of SAAP-148 were evaluated on only one clinical MRSA isolate here. However, the model can be extended to the investigation of further isolates including those of clinical relevance in device-associated infections such as Coagulase-negative staphylococci and enterococci. Of note, the effectiveness of SAAP-148 against Gram-positive and Gram-negative bacteria in mature biofilms has been confirmed in another study (Nibbering et al., personal communication). SAAP-148 formulated in an ointment was also highly effective against an established biofilm-associated infection on wounded *ex vivo* human skin models [10]. Together, SAAP-148 is the most promising peptide for further development as novel agent to combat biofilm-associated infections. In order to prevent PJI, a SAAP-148 formulation may be developed as a coating for prosthetic joints and/or for application to the tissues surrounding the implant. SAAP-148 could also be used as adjunctive treatment during surgical debridement to rapidly kill any surviving bacteria after debridement.

Other strategies to combat persisters

In addition to the application of antimicrobial peptides, mechanic or enzymatic disruption of the matrix of the biofilms may also prevent the subsequent awakening of persisters rendering them susceptible to antibiotics again. Other innovative strategies, like bacteriophages and heat induction, should be further explored as viable approaches to combat clinical device-associated infections [31 32]. The biofilm model described in this study is well suited to investigate the possibilities and limitations of these strategies in more detail. Finally, combinations of various strategies may have the largest clinical effect on biofilm-associated infections.

Conclusions

Novel *in vitro* models simulating PJI have been developed to evaluate the effects of antimicrobial peptides on persisters residing in antibiotics-exposed, mature biofilms. Combined rifampicin/ciprofloxacin treatment did not eliminate all biofilm-embedded bacteria, indicating the presence of persisters within mature biofilms. Microcalorimetry confirmed the dormant state of these bacteria. SAAP-148 eliminated persisters within mature biofilms on abiotic surfaces. SAAP-148 was more effective than LL-37, pexiganan and ADEP4. Based on these data, SAAP-148 is a promising candidate for further development as agent to treat patients suffering from biofilm-associated infections like PJI.

Acknowledgments

We thank dr. Willem van Wamel and MSc Valerie Baede (Dept of Medical Microbiology, Erasmus Medical Center, Rotterdam, the Netherlands) for their assistance with the heat flow measurements using microcalorimetry.

Conflicts of interests

The authors have no conflicts of interests to disclose.

Funding

No funding was obtained for this study

Meetings were the data were presented

European Bone and Joint Infection Society (EBJIS) Conference September 2019 Antwerp, Belgium

References

1. Tande AJ, Patel R. Prosthetic joint infection. *Clin Microbiol Rev* 2014;27(2):302-45. doi: 10.1128/CMR.00111-13 [published Online First: 2014/04/04]
2. Zeller V, Kerroumi Y, Meyssonnier V, et al. Analysis of postoperative and hematogenous prosthetic joint-infection microbiological patterns in a large cohort. *The Journal of infection* 2018 doi: 10.1016/j.jinf.2017.12.016 [published Online First: 2018/02/06]
3. Lora-Tamayo J, Murillo O, Iribarren JA, et al. A large multicenter study of methicillin-susceptible and methicillin-resistant Staphylococcus aureus prosthetic joint infections managed with implant retention. *Clin Infect Dis* 2013;56(2):182-94. doi: cis746 [pii];10.1093/cid/cis746 [doi]
4. Lora-Tamayo J, Senneville E, Ribera A, et al. The Not-So-Good Prognosis of Streptococcal Periprosthetic Joint Infection Managed by Implant Retention: The Results of a Large Multicenter Study. *Clin Infect Dis* 2017;64(12):1742-52. doi: 10.1093/cid/cix227 [published Online First: 2017/04/04]
5. Lewis K. Persister cells. *Annu Rev Microbiol* 2010;64:357-72. doi: 10.1146/annurev.micro.112408.134306 [doi]
6. Balaban NQ, Merrin J, Chait R, et al. Bacterial persistence as a phenotypic switch. *Science* 2004;305(5690):1622-5. doi: 10.1126/science.1099390 [published Online First: 2004/08/17]
7. Harms A, Maisonneuve E, Gerdes K. Mechanisms of bacterial persistence during stress and antibiotic exposure. *Science* 2016;354(6318) doi: 10.1126/science.aaf4268 [published Online First: 2016/12/17]
8. Conlon BP, Nakayasu ES, Fleck LE, et al. Activated ClpP kills persisters and eradicates a chronic biofilm infection. *Nature* 2013;503(7476):365-70. doi: nature12790 [pii];10.1038/nature12790 [doi]
9. van der Does AM, Beekhuizen H, Ravensbergen B, et al. LL-37 directs macrophage differentiation toward macrophages with a proinflammatory signature. *J Immunol* 2010;185(3):1442-9. doi: 10.4049/jimmunol.1000376 [published Online First: 2010/07/09]
10. de Breij A, Riool M, Cordfunke RA, et al. The antimicrobial peptide SAAP-148 combats drug-resistant bacteria and biofilms. *Science translational medicine* 2018;10(423) doi: 10.1126/scitranslmed.aan4044 [published Online First: 2018/01/13]
11. Ge Y, MacDonald DL, Holroyd KJ, et al. In vitro antibacterial properties of pexiganan, an analog of magainin. *Antimicrob Agents Chemother* 1999;43(4):782-8. [published Online First: 1999/04/02]
12. Nell MJ, Tjabringa GS, Wafelman AR, et al. Development of novel LL-37 derived antimicrobial peptides with LPS and LTA neutralizing and antimicrobial activities for therapeutic application. *Peptides* 2006;27(4):649-60. doi: 10.1016/j.peptides.2005.09.016 [published Online First: 2005/11/09]
13. Wolcott RD, Rumbaugh KP, James G, et al. Biofilm maturity studies indicate sharp debridement opens a time- dependent therapeutic window. *J Wound Care* 2010;19(8):320-8. doi: 10.12968/jowc.2010.19.8.77709 [published Online First: 2010/09/21]
14. Zimmerli W, Sendi P. Role of Rifampin against Staphylococcal Biofilm Infections In Vitro, in Animal Models, and in Orthopedic-Device-Related Infections. *Antimicrob Agents Chemother* 2019;63(2) doi: 10.1128/AAC.01746-18 [published Online First: 2018/11/21]
15. Braissant O, Wirz D, Gopfert B, et al. Use of isothermal microcalorimetry to monitor microbial activities. *FEMS Microbiol Lett* 2010;303(1):1-8. doi: 10.1111/j.1574-6968.2009.01819.x [published Online First: 2009/11/10]

16. Vulin C, Leimer N, Huemer M, et al. Prolonged bacterial lag time results in small colony variants that represent a sub-population of persisters. *Nat Commun* 2018;9(1):4074. doi: 10.1038/s41467-018-06527-0 [published Online First: 2018/10/06]
17. Lewis K, Shan Y. Persister Awakening. *Mol Cell* 2016;63(1):3-4. doi: 10.1016/j.molcel.2016.06.025 [published Online First: 2016/07/09]
18. Dunne WM, Jr., Mason EO, Jr., Kaplan SL. Diffusion of rifampin and vancomycin through a Staphylococcus epidermidis biofilm. *Antimicrob Agents Chemother* 1993;37(12):2522-26.
19. Reiter KC, Sambrano GE, Villa B, et al. Rifampicin fails to eradicate mature biofilm formed by methicillin-resistant Staphylococcus aureus. *Rev Soc Bras Med Trop* 2012;45(4):471-4. [published Online First: 2012/08/30]
20. Molina-Manso D, del Prado G, Ortiz-Perez A, et al. In vitro susceptibility to antibiotics of staphylococci in biofilms isolated from orthopaedic infections. *International journal of antimicrobial agents* 2013;41(6):521-3. doi: 10.1016/j.ijantimicag.2013.02.018 [published Online First: 2013/04/25]
21. Jorgensen NP, Skovdal SM, Meyer RL, et al. Rifampicin-containing combinations are superior to combinations of vancomycin, linezolid and daptomycin against Staphylococcus aureus biofilm infection in vivo and in vitro. *Pathog Dis* 2016;74(4):ftw019. doi: 10.1093/femspd/ftw019 [published Online First: 2016/04/03]
22. Kwan BW, Valenta JA, Benedik MJ, et al. Arrested protein synthesis increases persister-like cell formation. *Antimicrob Agents Chemother* 2013;57(3):1468-73. doi: 10.1128/AAC.02135-12 [published Online First: 2013/01/09]
23. Widmer AF, Frei R, Rajacic Z, et al. Correlation between in vivo and in vitro efficacy of antimicrobial agents against foreign body infections. *J Infect Dis* 1990;162(1):96-102.
24. Zimmerli W, Frei R, Widmer AF, et al. Microbiological tests to predict treatment outcome in experimental device-related infections due to Staphylococcus aureus. *J Antimicrob Chemother* 1994;33(5):959-67.
25. Trampuz A, Murphy CK, Rothstein DM, et al. Efficacy of a novel rifamycin derivative, ABI-0043, against Staphylococcus aureus in an experimental model of foreign-body infection. *Antimicrob Agents Chemother* 2007;51(7):2540-5. doi: 10.1128/AAC.00120-07 [published Online First: 2007/05/16]
26. Lucet JC, Herrmann M, Rohner P, et al. Treatment of experimental foreign body infection caused by methicillin-resistant Staphylococcus aureus. *Antimicrob Agents Chemother* 1990;34(12):2312-7. doi: 10.1128/aac.34.12.2312 [published Online First: 1990/12/01]
27. Tshefu K, Zimmerli W, Waldvogel FA. Short-term administration of rifampin in the prevention or eradication of infection due to foreign bodies. *Rev Infect Dis* 1983;5 Suppl 3:S474-S80.
28. Brotz-Oesterhelt H, Beyer D, Kroll HP, et al. Dysregulation of bacterial proteolytic machinery by a new class of antibiotics. *Nat Med* 2005;11(10):1082-7. doi: 10.1038/nm1306 [published Online First: 2005/10/04]
29. Conlon BP, Rowe SE, Gandt AB, et al. Persister formation in Staphylococcus aureus is associated with ATP depletion. *Nat Microbiol* 2016;1:16051. doi: 10.1038/nmicrobiol.2016.51 [published Online First: 2016/08/31]
30. Shan Y, Brown Gandt A, Rowe SE, et al. ATP-Dependent Persister Formation in Escherichia coli. *mBio* 2017;8(1) doi: 10.1128/mBio.02267-16 [published Online First: 2017/02/09]
31. Pijls BG, Sanders I, Kuijper EJ, et al. Segmental induction heating of orthopaedic metal implants. *Bone Joint Res* 2018;7(11):609-19. doi: 10.1302/2046-3758.711.BJR-2018-0080.R1 [published Online First: 2018/12/26]
32. Morris JL, Letson HL, Elliott L, et al. Evaluation of bacteriophage as an adjunct therapy for treatment of peri-prosthetic joint infection caused by Staphylococcus aureus. *PLoS One* 2019;14(12):e0226574. doi: 10.1371/journal.pone.0226574 [published Online First: 2019/12/27]

CHAPTER 8

Summary and general discussion

The road to improved outcome for patients with a prosthetic joint infection is challenging and requires multidisciplinary collaboration. The evidence for diagnostic and treatment strategies for PJI is scarce as demonstrated by the international consensus meeting in 2018 in the United States during which expert-based consensus about diagnosis and treatment was reached by voting. In The Netherlands, this lack of evidence is reflected by much practice variation between different PJI treatment centers, even at geographically adjacent hospitals. In 2015, orthopedic surgeons, infectious diseases specialists and medical microbiologists from several regional hospitals increasingly felt the need to develop scientific evidence by comparing the then used protocols for treatment of PJI. Further, we aimed to cooperate and harmonize the practice variation in our region. A diagnostic and treatment protocol was developed by all participating centers, weekly multidisciplinary meetings were organized and date were collected prospectively in a regional quality registry. This collaboration increased not only job satisfaction but resulted in a more scientific approach leading to important insights as summarized in this thesis. Next to the scientific evaluation of clinical treatment strategies, translational research is needed to understand the exact pathophysiologic mechanism of surviving persisters in a biofilm. Such knowledge charts the scientific route to innovative anti-persister treatment strategies. In this chapter, three key outcomes of the research, described in this thesis, will be summarized and discussed.

The use of E health to detect prosthetic joint infections

The *first* part focuses on the role of mobile E-health aimed at earlier detection of PJI. In **chapter 2**, the introduction of a postoperative woundcare app, which was developed to increase patient involvement and to shorten the time to PJI diagnosis, is described. In this study, we focused on assessing the ease of use and perceived usefulness of this app in a group of sixty-nine patients in two hospitals. The use of this app was evaluated by patients with a high perceived usefulness and ease of use. The patient-reported and physician-reported outcome were identical in 80% of cases. The high self-reported perceived usefulness and ease of use was the reason to set up a larger, multicenter study in which the same app was used to gain better insight in the duration and amount of wound leakage in patient who developed a PJI and in patients with an uncomplicated course. In **chapter 3**, the results of this study were summarized. From this study, it appeared that PJI was very unlikely in postoperative patients without any wound leakage or other signs of wound infection. Postoperative wound leakage in the first week after arthroplasty frequently occurred (50%) and was not related to PJI. Apparently, this early wound leakage can be regarded as a natural postoperative course which is only relevant it the wound continues to leak over the next weeks. Wound leakage in the second or third week however was strongly associated with the occurrence of PJI, but its positive predictive value was low. For example, any amount of wound leakage in the third

postoperative week was strongly associated with PJI (OR 51, 95% CI 11-227, sensitivity 88%, specificity 88%). However, the positive predicted value was only 11%, which would result in an unacceptable number of 10 patients needed to DAIR to diagnose and treat one real PJI. We estimate that host characteristics such as weight, presence of diabetes mellitus, and use of anticoagulants may lead to a longer duration of wound leakage rather than being a sign of early wound infection. In contrast, moderate to heavy leakage in the third postoperative week predicted PJI with much higher specificity (PPV 83%) resulting in a Number Needed to DAIR to diagnose one patient with PJI of 1.2 patients.

How should the result of this study influence daily practice? One consequence may be that outpatient wound care follow-up after arthroplasty may be reduced or even cancelled for patients who report no leakage and no other complications during the postoperative period. Outpatient follow up may still be needed for evaluation of mobility, strength and other functional tests which cannot be performed with telemonitoring. An important learning point is that early wound leakage after arthroplasty belongs to the natural postoperative course, except if it lasts longer than two weeks or increases in quantity. Even in the third postoperative week, mild leakage is still found in many patients with an uncomplicated course.

The wound care app was intended to improve patient engagement and prevent delay in the diagnosis of PJI. Many patients (42%) felt more involved due to using the app while less patients (15%) felt partly involved or not / only a bit involved (28%). We suspect that patients with an excellent outcome without complications are less in need of an app to feel more involved in care. Another question is whether the use of the app attributed to earlier diagnosis of complications. The median time-to-DAIR in our study was 16 days. Only 2 PJIs (13%) were diagnosed between week 4 and 12 after arthroplasty. These differential time-to-DAIR is short if compared to data from a recent Dutch study in which the differential time-to-DAIR for most patients was between four and twelve weeks after arthroplasty (56% of knee PJIs and 36% of hip PJIs). In a Swedish cohort, the differential time-to-DAIR was 20 days for patients with total hip arthroplasty.[1] Three of the six patients with an eventual PJI in our study were earlier admitted or seen at the outpatient clinic after an alert-based phone call to the hospital. This shows the app's potential to speed up the diagnosis of prosthetic infection. However, the small number of patients with PJI in our study necessitates cautious conclusions. A randomized controlled trial in which the time to DAIR for PJI is compared in patients with and without the use of a wound care app, would answer this question. The app used in this study specifically targeted wound leakage and signs of wound infection. Ideally, such an application should not be a standalone wound leakage app but a more general perioperative app in which all aspects of perioperative care for patients are integrated. Based on the current study, the algorithm should be adjusted to reduce unnecessary alerts to patients. The predictive value of the algorithm may be improved by using a machine

learning algorithm, where changes can be made to the algorithm automatically, based on mounting collected data. Adding laboratory parameters like an increase in C-reactive protein may also increase the yield of the algorithm. Based on the current study, less value should be placed on minimal wound leakage and low pain scores, as these were not discriminatory for the development of PJI.

Evaluation of current antimicrobial strategies for PJI

The *second* part of this thesis focuses on the evaluation of different antimicrobial treatment strategies for PJI. In **chapter 4** all studies reporting the outcome of staphylococcal PJI after DAIR over the last 30 years were assessed in a systematic review and meta-analysis, focused on the use of rifampicin for staphylococcal PJI. One of the conclusions of this study was the persistently low success rates after DAIR, although there was a trend toward increasing success rates over the years. The added value of rifampicin, compared with other treatment strategies for staphylococcal prosthetic joint infections, appeared to be marginal. The trim-and-fill analysis done in our systematic review suggested publication bias. Correction for this bias resulted in an adjusted relative risk of success of 1.04 (95%CI 0.94 to 1.14) when rifampicin was used.

Despite the limited evidence for the effectiveness of rifampicin, the recommendations to use rifampicin in staphylococcal prosthetic joint infections are strong in most guidelines. Several explanations for this can be given. The study by Zimmerli et al., in which almost all patients recovered after treatment with DAIR and combination treatment with rifampicin, was published in 1998. At that time the outcome after DAIR for PJI was regarded as poor, although large cohort studies before 1998 do not exist. Although the trial was heavily underpowered, including only 18 patients with a PJI, the good outcome in the group treated with rifampicin was in line with several experimental foreign body animal models showing high cure rates if rifampicin combination treatment was used. Meanwhile, the use of rifampicin is widely implemented in the care of patients with infected implants. Our systematic review, described in **chapter 4**, was the first review that systematically appraised all studies regarding the outcome of staphylococcal PJI. The methodological quality of most observational studies in which the use of rifampicin for PJI was evaluated, was poor. To initiate scientific discussion on methodological limitations in observational studies on rifampicin, we wrote two letters to the editor. In these letters, we drew attention to the various forms of bias and confounding in observational studies on PJI. In **chapter 4** we contested the conclusions of an observational study in which authors concluded that prolonged duration of rifampicin therapy was a key determinant for improved outcomes in acute staphylococcal PJI treated with DAIR. However, this outcome may be explained by (1) exclusion of patients who failed during treatment with rifampicin (exclusion bias) , (2) confounding by indication by not prescribing rifampicin

to patients with a higher apriori risk of therapy failure and (3) immortal time bias by prescribing rifampicin only to patients who did not have treatment failure in the first postoperative weeks after the DAIR. In **chapter 4**, we also discussed the study by Beldman et al, who demonstrated a statistically significant association between rifampicin use and treatment success in pooled cohorts[2] However, confounding and immortal time bias were still likely to be present and cannot be fully adjusted for in a multivariate analysis. An important limitation of observational PJI studies is the comparison between rifampicin and non-rifampicin treatment strategies. Comparison of one well-defined strategy (rifampicin combination therapy) with all other nón-defined strategies (including all varieties of antimicrobial options without rifampicin) will, together with the forms of bias described above, likely result in an underestimation of the effectiveness of all regimens included in the non-rifampicin strategies. This bias in favor of a well-defined treatment strategy may lead to unjustified rejection of equally good alternatives within the non-defined treatment group. This is further elaborated under the next heading.

The risk of PJI in patients undergoing endoprosthetic reconstruction of the lower extremities after tumor surgery is high and often requires multiple surgical interventions. In **chapter 5** we focused on outcome after surgical debridement for patients with an infected megaprosthesis after tumour surgery. We found more polymicrobial infections in these patients compared to PJI after conventional arthroplasty. This is in line with an earlier study by our group in which 25% of patients had polymicrobial PJI.[3] The success rate of DAIR for an infected megaprosthesis was 50%. The chance of eradicating the infection after each subsequent DAIR was approximately 30-50%. This low success rate may be related to the chronicity of infections (35% had a DAIR for chronic PJI more than 12 weeks after index surgery), a known risk factor for failure after DAIR. Complete exchange of the megaprosthesis when infected may enhance cure rates but a much more complicated surgical procedure is needed for this strategy. Practically, it also takes more time to construct a new custom-made tumour prosthesis, making a one-stage exchange of acutely infected tumour prostheses more challenging. Weighing all these arguments, performing one or more DAIR procedures appears to be a viable treatment option for patients for whom there is no contraindication to DAIR.

New antimicrobial strategies for PJI: antibiotic treatment

To better understand the group of patients not treated with rifampicin, we analyzed data from patients with staphylococcal PJI treated with alternative strategies. In **chapter 6**, we analyzed data from 200 patients with staphylococcal PJI in our prospective observational clinical registry. In this group of patients, clindamycin-based treatment was found to be more effective than non-clindamycin-based treatment, but for the same group of patients, rifampicin-based treatment was also more effective than non-rifampicin-based treatment (Figure 1).

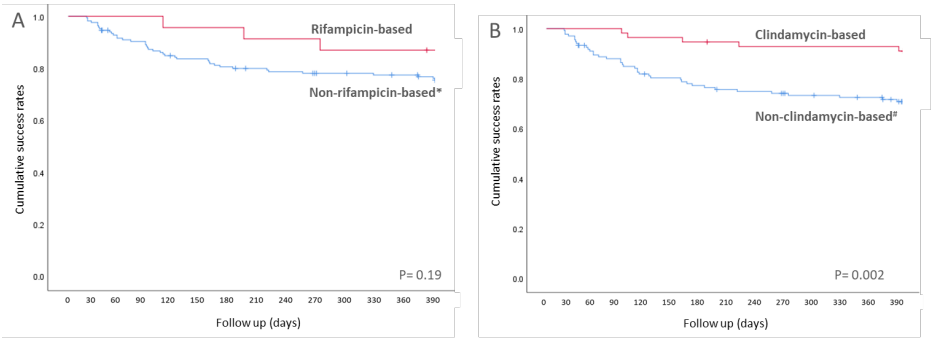


Figure 1. Outcome after DAIR related to antimicrobial strategy

This analysis clearly demonstrates the limitation of comparing treatment strategies if one of the group is poorly defined. To overcome this issue, we classified patients in several well-defined antimicrobial treatment groups. This enabled us to draw relevant conclusions about the effectivity of different treatment regimens. A treatment strategy with either clindamycin or flucloxacillin and only five days of rifampicin was found to be as effective as traditional long-term rifampicin combination therapy. This non-inferiority was achieved even at a four-week shorter treatment duration in the patients treated with flucloxacillin or clindamycin. The results of this study are in line with the results of an earlier report, also described in **Chapter 6.** Here, the use of targeted oral flucloxacillin monotherapy for staphylococcal PJI is reported in a small observational cohort study. A success rate of 83% is reported in patients with staphylococcal hip PJI and 44% in staphylococcal knee PJI treated with flucloxacillin monotherapy and only five days of rifampicin, started immediately postoperative**.** Both studies are the first reports indicating that reasonable cure rates can be achieved with alternative targeted strategies with antimicrobial monotherapy. However, confounding is also present in this study and groups. The groups were well defined but not always comparable. Clindamycin, for example, was only prescribed at the time of the iv-oral switch. Patients in this group probably had a more favorable prognosis than those for whom it was deemed necessary to continue treatment with intravenous flucloxacillin and who were therefore assigned to the flucloxacillin group. This can only be solved with randomisation. Therefore, based on the findings described in this thesis, a randomized controlled trial is needed to directly compare clindamycin or flucloxacillin monotherapy with rifampicin combination therapy.

In the past 30 years, two randomized studies have been conducted to answer this question. The first study, published in 1998, showed a significantly better outcome when rifampicin was used but failure in the control group was mainly caused by ciprofloxacin resistance and the study included only 18 patients with prosthetic joint infection[4]. A larger, more

recent study including 48 patients was published in 2020 and showed no difference between treatment with or without rifampicin[5]. Both studies were underpowered due to low inclusion rates. Therefore, the most recent study did not lead to a change in guidelines. The lack of good evidence, the disadvantages of long-term combination therapy with rifampicin and fluoroquinolones, the toxicities and drug-drug interactions associated with the use if rifampicin and the need for equivalent treatment alternatives justify the set up of a new trial. Therefore, in 2023, a multicenter study will start in the Netherlands in which patients will be randomized between clindamycin monotherapy and rifampicin/levofloxacin combination therapy during the oral treatment phase of prosthetic joint infections caused by staphylococci (**Ri**fampicin **Co**mbination **T**herapy versus **T**argeted **A**ntimicrobial Monotherapy in the oral antimicrobial treatment phase of staphylococcal prosthetic joint infection; the RiCOTTA trial).

There are few clinical data about the optimal timing of starting rifampicin in the treatment of PJI. Treatment with rifampicin may result in selection of rifampicin-resistant coagulase-negative staphylococci on the skin that could potentially infect the prosthesis via the postoperative wound and cause a secondary superinfection of the prosthesis. However, clinical data supporting this risk are lacking, and withholding an adequate bactericidal agent because of a possible complication seems illogical. In the two cohort studies in this thesis (**chapter 6**), in which rifampicin was started immediately postoperative, only one patient developed a relapse with a rifampicin-resistant *S. aureus*, one year after the DAIR. Because of the time elapsed since surgery, this resistance was probably not related to the five days of rifampicin treatment one year before. In the randomized controlled trial by Zimmerli et al., rifampicin was also started immediately postoperatively and did not result in rifampicin-resistant staphylococci in patients with a relapse.[4] In addition, in vitro studies show that rifampicin resistance only develops under the condition of a high bacterial load and if rifampicin is given as monotherapy. During DAIR, the bacterial load is significantly reduced intraoperatively and rifampicin is always started as combination therapy. Based on all these data, we therefore consider immediate postoperative initiation of rifampicin safe

New antimicrobial strategies for PJI: anti-persister treatment

Innovative strategies to eradicate biofilm-embedded bacteria are the focus of the *third* part of this thesis. Persister cells residing within chronic biofilms are the root cause of relapse of biofilm-associated infections because they cannot be targeted by antibiotics. In **chapter 7**, we confirmed the antibiotic recalcitrance of biofilms by demonstrating that highly bactericidal antibiotics (rifampicin combined with ciprofloxacin) were not able to eradicate persisters within a mature biofilm. Therefore, the development of alternative anti-persister drugs is necessary to cure biofilm-associated infections. If such a drug

would be available, surgery for PJI and many other biofilm-associated infections like vascular graft infections, prosthetic valve endocarditis, fracture-related infections, spinal implant infections and infected cardiac devices may be no longer needed. Unfortunately, the global preclinical antibacterial pipeline does not include any anti-persister drug. However, several anti-persister treatment strategies have been developed or the last decades which may lead to clinical application in the future. Antimicrobial peptides have broad antibacterial activities and have shown activity against persisters.[6] SAAP-148 is an antimicrobial peptide, developed at LUMC, which is effective under physiological conditions (i.e., in 50% human plasma) and has broad antimicrobial activity against MRSA and Gram-negative bacteria in ex vivo and in vivo wound infections.[6] We decided to optimize preclinical research models with chronic biofilms on abiotic surfaces to test anti-persister drugs. In **chapter 7**, we report on the development of an in vitro mature biofilm model. With this model we aimed to develop an innovative way to simulate a PJI as much as possible, creating optimal conditions to create a mature biofilm as is the case in patients with a PJI. With this approach we tried to avoid outcomes of in vitro experiments that may not be optimal for translation to clinical biofilm-associated infections. We assessed the effectivity of anti-biofilm and anti-persister agents on polystyrene plates, titanium/aluminium/niobium discs and prosthetic joint liners. Bacteria obtained from and residing within these biofilms were eradicated after exposure to SAAP-148, acyldepsipeptide-4, LL-37 and pexiganan. SAAP-148 also eradicated bacteria within the antibiotic-exposed, mature biofilms on all surfaces, indicating that SAAP-148 is highly effective against persisters within these models. This mature biofilms on different abiotic surfaces can be further used to test other novel treatment strategies like bacteriophages, quorum sensing inhibitors and other antimicrobial peptides. Application of SAAP-148 in an ointment solution on an infected implant as additional treatment during surgical debridement would be a relevant clinical application which needs further investigation.

Concluding remarks

Accurate self-monitoring of postoperative wounds after joint implantation helped elucidate the course of wound leakage and its association with acute prosthetic joint infections. The collection of clinical data on different antimicrobial treatment strategies provided insight into the effectiveness of different treatment options for patients with a prosthetic joint infection. In this thesis, we report that personalized antimicrobial treatment for prosthetic joint infections is possible without compromising the effectiveness of treatment. In the coming years, the role of different oral treatment strategies will be further studied in the already mentioned multicenter RiCOTTA study in the Netherlands. In addition, this thesis describes the role and importance of new anti-persister drugs against biofilm-associated infections. We developed a biofilm model that closely resembles the clinic of a prosthetic joint infection. This allowed us to investigate the effectiveness of innovative anti-biofilm drugs. Based on the results described in this thesis, future research will be aimed at better understanding the pathogenesis of biofilms. The effectiveness of new drugs against biofilms can be investigated in this biofilm model. This should ultimately lead to better treatment options for patients with a prosthetic joint infection, ultimately achieving the goal: better care for vulnerable patients who are confronted with a serious postoperative complication.

References

1. Svensson K, Rolfson O, Naucler E, et al. Exchange of Modular Components Improves Success of Debridement, Antibiotics, and Implant Retention: An Observational Study of 575 Patients with Infection After Primary Total Hip Arthroplasty. *JB JS Open Access* 2020;5(4) doi: 10.2106/JBJS.OA.20.00110 [published Online First: 2020/12/31]
2. Beldman M, Lowik C, Soriano A, et al. If, when, and how to use rifampin in acute staphylococcal periprosthetic joint infections, a multicentre observational study. *Clin Infect Dis* 2021 doi: 10.1093/cid/ciab426 [published Online First: 2021/05/11]
3. Sanders PTJ, Bus MPA, Scheper H, et al. Multiflora and Gram-Negative Microorganisms Predominate in Infections Affecting Pelvic Endoprostheses Following Tumor Resection. *J Bone Joint Surg Am* 2019;101(9):797-803. doi: 10.2106/JBJS.18.00836 [published Online First: 2019/05/03]
4. Zimmerli W, Widmer AF, Blatter M, et al. Role of rifampin for treatment of orthopedic implant-related staphylococcal infections: a randomized controlled trial. Foreign-Body Infection (FBI) Study Group. *JAMA* 1998;279(19):1537-41. doi: joc71805 [pii]
5. Karlsen OE, Borgen P, Bragnes B, et al. Rifampin combination therapy in staphylococcal prosthetic joint infections: a randomized controlled trial. *J Orthop Surg Res* 2020;15(1):365. doi: 10.1186/s13018-020-01877-2 [published Online First: 2020/08/30]
6. de Breij A, Riool M, Cordfunke RA, et al. The antimicrobial peptide SAAP-148 combats drug-resistant bacteria and biofilms. *Science translational medicine* 2018;10(423) doi: 10.1126/scitranslmed.aan4044 [published Online First: 2018/01/13]

Nederlandse samenvatting

De route naar een betere uitkomst voor patiënten met een geïnfecteerde gewrichtsprothese (prosthetic joint infection, PJI) is uitdagend en vereist multidisciplinaire samenwerking. Er wordt veel onderzoek verricht naar de beste strategieën voor diagnostiek en behandeling van PJI. Kwalitatief goede data zijn essentieel om goede strategieën te ontwikkelen. Dit blijkt bijvoorbeeld uit een internationale consensusbijeenkomst in de Verenigde Staten in 2018 waarbij op basis van stemming consensus werd bereikt over de diagnose en behandeling van prothese infecties. In Nederland wordt dit gebrek aan overtuigende data geïllustreerd door veel praktijkvariatie tussen verschillende PJI-behandelcentra. In 2015 besloot een groep orthopedisch chirurgen, internist-infectiologen en medisch microbiologen meer wetenschappelijk bewijs te verzamelen voor de in hun regio gebruikte protocollen voor de behandeling van prothese infecties. Daarnaast was het doel om de samenwerking te verbeteren en de praktijkvariatie in de regio te verminderen. De deelnemende centra ontwikkelden een protocol voor diagnostiek en behandeling. Er werden wekelijks multidisciplinaire bijeenkomsten georganiseerd in elk centrum en er werden prospectief data verzameld in een regionaal kwaliteitsregister. Deze samenwerking leidde tot de inzichten die samengevat zijn in dit proefschrift. Naast de wetenschappelijke evaluatie van in de klinische praktijk gebruikte strategieën voor behandeling is onderzoek nodig om het pathofysiologische mechanisme van overlevende bacteriën in een biofilm beter te begrijpen. Dit helpt om de wetenschappelijke route te bepalen naar vernieuwende behandelstrategieën waarmee bacteriën in biofilms gedood kunnen worden. In deze samenvatting worden de belangrijkste resultaten van het in dit proefschrift beschreven onderzoek besproken.

EHealth bij verdenking op een geïnfecteerde gewrichtsprothese

Het eerste deel van dit proefschrift beschrijft het gebruik van EHealth om PJI sneller te kunnen diagnosticeren en om informatie over de wond te verzamelen. In **hoofdstuk 2** wordt de introductie van een postoperatieve wondzorgapp beschreven, die werd ontwikkeld om de betrokkenheid van de patiënt te vergroten en de tijd tot de diagnose PJI te verkorten. We richtten ons hierin op het beoordelen van het gebruiksgemak en het ervaren nut van deze app in een groep van 69 patiënten. De app werd door patiënten beoordeeld met een hoge score voor ervaren nut en voor gebruiksgemak. Daarnaast kwamen de door patiënt en arts gerapporteerde uitkomsten in 80% van de gevallen overeen. Deze uitkomst was de reden om een grotere, multicenter studie op te zetten waarin dezelfde app werd gebruikt om beter inzicht te krijgen in de duur en de hoeveelheid wondlekkage bij patiënten die wel of geen PJI ontwikkelden. In **hoofdstuk 3** zijn de resultaten van deze studie samengevat. Het bleek dat een PJI zeer onwaarschijnlijk is bij patiënten bij wie na de operatie geen enkele wondlekkage optrad. Wondlekkage

in de eerste week na plaatsing van een gewrichtsprothese kwam vaak voor (50%). Deze wondlekkage was niet geassocieerd met het optreden van een prothese infectie. Blijkbaar kan deze vroege wondlekkage beschouwd worden als een natuurlijke postoperatief beloop. Wondlekkage in de 2e of 3e week was wel sterk geassocieerd met het optreden van PJI, maar trad ook op bij patiënten die geen PJI ontwikkelden en was geen goede voorspeller voor PJI. Wondlekkage in de derde week na operatie was sterk geassocieerd met PJI (sensitiviteit 88%, specificiteit 88%). De positief voorspellende waarde was slechts 11%, wat betekent dat er van de 10 patiënten die geopereerd zouden worden vanwege verdenking PJI, slechts één patiënt daadwerkelijk een prothese infectie heeft. Dit is een onacceptabel hoog aantal aan onterecht geopereerde patiënten. Wij denken dat andere factoren zoals overgewicht, suikerziekte en het gebruik van antistolling ook kunnen leiden tot een langere duur van ongecompliceerde wondlekkage zonder direct een teken te zijn van een vroege prothese infectie. Als een patiënt in de derde week na de plaatsing van de prothese matige tot forse wondlekkage had was dat wel een hele goede voorspeller voor een PJI (positief voorspellende waarde 83%). Dit komt statistisch gezien neer op het opereren van slechts 1,2 patiënten om één patiënt met PJI te behandelen.

Welke consequenties hebben deze resultaten voor de dagelijkse praktijk? De poliklinische wondzorg na plaatsing van een gewrichtsprothese zou kunnen worden verminderd of zelfs afgeschaft bij patiënten zonder lekkage of andere in de app gerapporteerde complicaties. Poliklinische follow-up kan nog wel nodig zijn voor evaluatie van mobiliteit, kracht en andere functionele testen die niet met telemonitoring kunnen worden uitgevoerd. Een ander leerpunt van deze studie is dat vroege wondlekkage na prothese plaatsing hoort bij het normale postoperatieve beloop, tenzij dit langer dan twee weken duurt of in hoeveelheid toeneemt. Zelfs in de derde postoperatieve week wordt bij veel patiënten nog steeds milde lekkage gevonden zonder dat zij later een prothese infectie ontwikkelen.

Een ander doel van de wondzorgapp was om de betrokkenheid van patiënten te verbeteren en vertraging in het diagnosticeren van PJI te voorkomen. Veel patiënten (42%) voelden zich meer betrokken dankzij het gebruik van de app, terwijl anderen (15%) zich gedeeltelijk of slechts een beetje of zelfs niet betrokken voelden (28%). We vermoeden dat patiënten met een uitstekende uitkomst zonder complicaties de app minder nodig hebben om zich betrokken te voelen bij de zorg. Een andere vraag is of het gebruik van de app heeft geleid tot eerder stellen van de diagnose prothese infectie. De mediane tijd van plaatsing prothese tot DAIR in onze studie was 16 dagen. Slechts 2 PJI's (13%) werden tussen week 4 en 12 na plaatsing van de prothese gediagnosticeerd. Deze tijd tot uitvoer van DAIR is vrij kort als je dat vergelijkt met gegevens uit een recente Nederlandse studie, waarin bij 56% van de patiënten met een knieprothese en 36% van de patiënten met een heupprothese de DAIR werd uitgevoerd tussen 4 en 12 weken. In

een Zweeds cohort was de tijd tot uitvoer van DAIR 20 dagen (bij patiënten met een heupprothese). In onze studie werden drie van de zes patiënten op basis van een alert via de app eerder opgenomen of op de polikliniek teruggezien met een PJI. Dit toont de potentie van de app om het diagnosticeren van een prothese infectie te bespoedigen. Door het geringe aantal patiënten met een prothese infectie in onze studie kan deze conclusie echter niet met zekerheid getrokken worden. Een gerandomiseerde studie waarin de tijd tot DAIR wordt vergeleken bij patiënten met en zonder het gebruik van een wondzorg app zou deze vraag beantwoorden.

De in deze studie gebruikte app was specifiek gericht op wondlekkage en tekenen van wondinfectie. Idealiter zou een dergelijke app niet als wondlekkage-app gebruikt moeten worden, maar geïntegreerd moeten worden in een algemene wondzorg app waarin alle aspecten van de zorg voor patiënten rondom de operatie worden meegenomen. Gebaseerd op de huidige studie moet het algoritme dan wel worden aangepast om onnodige waarschuwingen aan patiënten zoveel mogelijke te verminderen. De positief voorspellende waarde van het algoritme voor infecties kan worden verbeterd met gebruik van *machine learning*, waarbij automatisch wijzigingen aan het algoritme kunnen worden aangebracht op basis van verzamelde gegevens. Ook het toevoegen van laboratoriumparameters zoals C-reactief proteïne kan het rendement van het algoritme verhogen. Op basis van de huidige studie moet minder waarde worden toegekend aan minimale wondlekkage en lage pijnscores, aangezien deze niet onderscheidend waren voor het ontwikkelen van een PJI.

Evaluatie van de huidige antimicrobiële strategieën voor PJI

Het tweede deel van dit proefschrift richt zich op de evaluatie van verschillende antimicrobiële behandelstrategieën voor prothese infecties. In **hoofdstuk 4** worden alle studies die de uitkomst van stafylokokken PJI na DAIR in de afgelopen 30 jaar rapporteerden, beoordeeld in een systematische review en meta-analyse, gericht op het gebruik van rifampicine voor stafylokokken PJI. Een van de opvallende bevindingen van deze studie was het aanhoudend lage succespercentage na DAIR, hoewel er over de jaren wel een trend was naar stijging van succespercentages. Het gebruik van rifampicine voor stafylokokken PJI is frequent geëvalueerd in observationele studies; de klinische meerwaarde van rifampicine, vergeleken met andere behandelstrategieën voor stafylokokken prothese infecties, bleek marginaal. Uit analyse bleek ook dat er waarschijnlijk meer studies met gunstige resultaten dan studies met minder gunstige resultaten voor rifampicine gepubliceerd zijn (publicatiebias). Als je daarvoor corrigeert met een statistische test (*trim-and-fill* analyse) dan verdwijnt de klinische meerwaarde van rifampicine geheel.

De aanbeveling om rifampicine te gebruiken bij prothese infecties veroorzaakt door stafylokokken PJI is sterk in de meeste richtlijnen ondanks het beperkte bewijs voor de effectiviteit hiervan. Hiervoor zijn meerdere verklaringen mogelijk. In de studie van Zimmerli en anderen, gepubliceerd in 1998, genazen bijna alle patiënten na behandeling met rifampicine combinatietherapie na een DAIR (Debridement, Antibiotics and Implant Retention; dat is de chirurgische behandeling waarbij de prothese grondig wordt gereinigd tijdens de operatie en de patiënt vervolgens met antibiotica wordt nabehandeld). Echter, slechts 18 patiënten in deze studie hadden een PJI. In de groep patiënten die werd behandeld met alleen ciprofloxacine trad bij bijna alle patiënten tijdens de behandeling resistentie op tegen ciprofloxacine. Dit verklaart het grote verschil in uitkomsten tussen de twee groepen. In die tijd werden de kansen op genezing na DAIR bij prothese infecties als laag ingeschat, hoewel er geen publicaties zijn van grotere studies vóór 1998. Het goede resultaat in de rifampicine groep leidde daarom tot toenemend voorschrijven van rifampicine voor PJI. Daarnaast waren de resultaten in overeenstemming met verschillende experimentele diermodellen met geïnfecteerd kunstmateriaal waarin combinatiebehandeling met rifampicine ook leidde tot hoge genezingspercentages.

Tegenwoordig wordt rifampicine breed toegepast in de zorg voor patiënten met geïnfecteerd kunstmateriaal. De systematische review, beschreven in **hoofdstuk 4**, beoordeelde voor het eerst alle studies van de laatste 20 jaar waarin de uitkomst van stafylokokken prothese infecties na een DAIR werden geëvalueerd. De methodologische kwaliteit van de meeste observationele studies bleek laag. Om een wetenschappelijke discussie op gang te brengen over methodologische beperkingen in observationele studies over rifampicine, schreven wij twee ingezonden brieven. Hierin vroegen wij aandacht voor de verschillende vormen van methodologische beperkingen in observationele studies over PJI. In **hoofdstuk 4** betwisten wij de conclusies van een observationele studie waarin het langer behandelen met rifampicine een bepalende factor was voor betere resultaten bij acute stafylokokken prothese infecties na een DAIR. Deze uitkomst kan echter worden verklaard door (1) het selectief niet analyseren van patiënten in de rifampicine groep die faalden tijdens de behandeling (*exclusiebias*), (2) het niet voorschrijven van rifampicine aan patiënten met een hogere voorafkans op falen van behandeling (*confounding by indication)* en (3) het pas voorschrijven van rifampicine aan patiënten die in de eerste weken na de DAIR geen falen van de behandeling hadden (*immortal time bias)*. In **hoofdstuk 4** bespraken wij ook de studie van Beldman en anderen waarin een statistisch significante associatie tussen het gebruik van rifampicine en succes werd aangetoond. Ook in deze studie waren *confounding by indication* en *immortal time bias* waarschijnlijk nog steeds aanwezig. Ook met een multivariate analyse kan voor deze bias niet volledig worden gecorrigeerd. Een

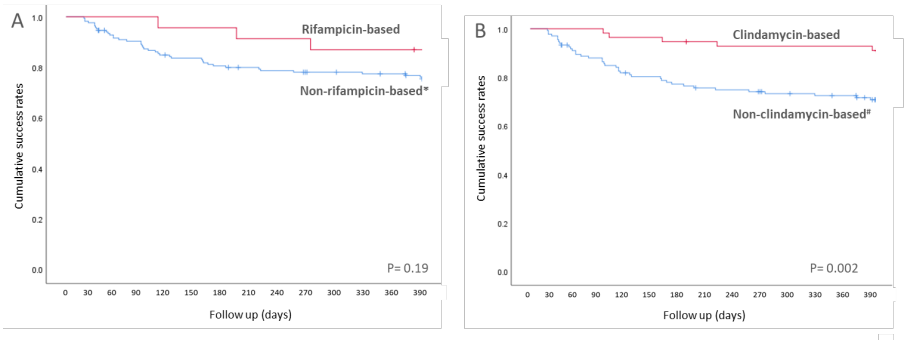
belangrijke methodologische beperking van de observationele studies is de vergelijking tussen rifampicine- en niet-rifampicine behandelstrategieën. Vergelijking van één goed gedefinieerde strategie (combinatiebehandeling met rifampicine) met alle andere niet-gedefinieerde strategieën bij elkaar (alle varianten van antimicrobiële therapie zonder rifampicine) zal, samen met de hierboven beschreven vormen van bias, waarschijnlijk leiden tot een onderschatting van effectieve behandelopties binnen de verzamelgroep van 'niet-rifampicine' strategieën. Dit kan leiden tot het ten onrechte verwerpen van goede alternatieven binnen de niet-gedefinieerde behandelingsgroep. Dit wordt verder aangetoond in de volgende paragraaf.

Het risico op een prothese infectie bij patiënten die na tumorchirurgie een grote gewrichtsreconstructie van de onderste extremiteiten ondergaan is hoog. In **hoofdstuk 5** richten wij ons op de uitkomst na DAIR van patiënten met een geïnfecteerde megaprothese na tumorchirurgie. Wij vonden meer polymicrobiële infecties bij deze patiënten in vergelijking met PJI na plaatsing van een conventionele gewrichtsprothese. Dit komt overeen met een eerdere studie van onze onderzoeksgroep waarin bij 25% van de patiënten de PJI veroorzaakt werd door meerdere verwekkers tegelijkertijd.

Het succespercentage van DAIR bij patiënten met een geïnfecteerde megaprothese was 50%. De kans op genezing na elke volgende DAIR was ongeveer 30-50%. Dit lage succespercentage kan verband houden met de chroniciteit van de infecties (35% had een DAIR voor chronische PJI, meer dan 12 weken na de indexoperatie), wat een bekende risicofactor is voor het falen na DAIR. Volledige vervanging van de megaprothese bij infectie kan het genezingspercentage verhogen, maar voor deze strategie is een gecompliceerdere chirurgische procedure nodig. Dit heeft onder andere te maken met de tijd die het kost om een nieuwe, op maat gemaakte tumorprothese te maken. Vervanging van acuut geïnfecteerde tumorprothesen in een enkele chirurgische procedure is daarom vaak niet mogelijk. Alles afwegend lijkt het uitvoeren van één of meerdere debridements een haalbare behandeloptie voor patiënten die in staat zijn een DAIR te ondergaan.

Nieuwe antimicrobiële strategieën voor PJI: behandeling met antibiotica

Wat zijn geschikte antimicrobiële opties voor patiënten met een prothese infectie door stafylokokken? Om deze vraag te beantwoorden hebben wij de gegevens geanalyseerd van patiënten met een stafylokokken PJI in onze regio. In **hoofdstuk 6** analyseerden wij gegevens van 200 patiënten met stafylokokken PJI uit een prospectieve observationele kwaliteitsregistratie. In dit cohort bleek clindamycine-gebaseerde behandeling effectiever dan niet-clindamycine-gebaseerde behandeling, maar in hetzelfde cohort was ook rifampicine-gebaseerde behandeling effectiever dan niet-rifampicine-gebaseerde behandeling (Figuur 1).



Figuur 1. Uitkomsten na DAIR gerealteerd aan antimicrobiele strategie

Deze analyse toont de beperking aan van het vergelijken van behandelstrategieën als groepen niet goed gedefinieerd zijn. In de groep patiënten die niet met rifampicine behandeld werd zaten subgroepen van patiënten met goed gedefinieerde behandelingen (clindamycine, flucloxacilline en vancomycine) Door deze patiënten vooraf in te delen in duidelijk gedefinieerde behandelgroepen kon de effectiviteit van de verschillende behandelregimes met elkaar vergeleken worden. Uitbehandeling met clindamycine of flucloxacilline was in deze studie even effectief als langdurige combinatietherapie met rifampicine en ciprofloxacine.

Deze vergelijkbare effectiviteit van flucloxacilline of clindamycine werd zelfs bereikt bij een vier weken kortere behandelingsduur. De resultaten van deze studie zijn in overeenstemming met de resultaten van een eerder rapport, dat ook in **hoofdstuk 6** wordt beschreven en waarin gerichte behandeling met oraal flucloxacilline resulteerde in een succespercentage van 83% bij patiënten met stafylokokken heup PJI en 44% bij stafylokokken knie PJI. Ook in deze studie werden patiënten behandeld met slechts vijf dagen rifampicine, wat direct postoperatief werd gestart.

Kunnen we uit deze studies concluderen dat alternatieve behandelstrategieën even effectief zijn als langdurige rifampicine combinatiebehandeling? Ook in de hier beschreven studie speelt confounding echter een rol. De groepen waren wel goed gedefinieerd maar niet altijd goed vergelijkbaar. Clindamycine werd bijvoorbeeld pas voorgeschreven bij de switch van intraveneuze naar orale therapie. Patiënten in deze groep hadden waarschijnlijk een gunstiger prognose dan de patiënten voor wie het nodig werd geacht de behandeling met intraveneus flucloxacilline langer te continueren en die daardoor in de flucloxacilline groep werden ingedeeld. Dit kan alleen opgelost worden met randomisatie.

In de afgelopen 30 jaar zijn twee gerandomiseerde studies verricht om deze vraag te beantwoorden. De eerste studie uit 1998 is reeds besproken in deze samenvatting. Een recentere en grotere, gerandomiseerde studie met 48 patiënten, gepubliceerd in 2020, toonde geen verschil tussen behandeling met of zonder rifampicine. Ook deze studie had helaas onvoldoende bewijskracht vanwege het lage aantal geïncludeerde patiënten. De meest recente studie heeft daarom niet tot een verandering van richtlijnen geleid. Het gebrek aan goede bewijskracht, de nadelen van langdurige combinatietherapie met rifampicine en fluorochinolonen en de behoefte aan gelijkwaardige behandelalternatieven rechtvaardigen daarom het uitvoeren van een nieuwe gerandomiseerde studie waarin gerichte monotherapie rechtstreeks vergeleken wordt met rifampicine combinatietherapie. In 2023 start in Nederland daarom een multicenter studie waarin patiënten zullen worden gerandomiseerd tussen clindamycine monotherapie en rifampicine/levofloxacine combinatietherapie tijdens de orale behandelfase van prothese infecties veroorzaakt door stafylokokken (**Ri**fampicin **Co**mbination **T**herapy versus **T**argeted **A**ntimicrobial Monotherapy in the oral antimicrobial treatment phase of staphylococcal prosthetic joint infection; de RiCOTTA-studie).

Voor de optimale timing van het starten van rifampicine bij de behandeling van PJI zijn weinig klinische data bekend. Behandeling met rifampicine kan leiden tot selectie van rifampicineresistente Coagulase-negatieve stafylokokken op de huid die mogelijk via de postoperatieve wond de prothese kunnen infecteren en een secundaire superinfectie van de prothese kunnen veroorzaken. Klinische gegevens die de zorg om dit risico ondersteunen ontbreken echter. Daarnaast lijkt het onthouden van een adequaat bacteriedodend middel aan een patiënt vanwege een mogelijke complicatie onlogisch. In de twee hierboven samengevatte studies, waarin direct na de operatie gestart werd met rifampicine, ontwikkelde slechts één patiënt een recidief met een rifampicine resistente *S. aureus*, een jaar na de DAIR. Gezien het lange tijdsinterval tussen operatie en re-infectie had resistentie waarschijnlijk geen verband met de vijf dagen rifampicinebehandeling van een jaar terug. In de gerandomiseerde gecontroleerde studie van Zimmerli et al. werd rifampicine ook onmiddellijk postoperatief gestart en resulteerde dit niet in rifampicineresistente stafylokokken bij patiënten die een recidief ontwikkelden. Bovendien blijkt uit in vitro-onderzoek dat rifampicineresistentie zich alleen ontwikkelt bij een hoge bacteriële load *en* als rifampicine als monotherapie wordt voorgeschreven. Tijdens een DAIR wordt de bacteriële load tijdens de operatie aanzienlijk verminderd *en* wordt rifampicine gestart in combinatie met een tweede antibioticum. Op basis van al deze gegevens achten wij onmiddellijke postoperatieve start van rifampicine dan ook veilig.

Nieuwe antimicrobiële strategieën voor PJI: behandeling tegen persisters

Vernieuwende strategieën om bacteriën binnenin een biofilm te doden staan centraal in het derde deel van dit proefschrift. Persisters in chronische biofilms zijn de belangrijkste oorzaak van het falen van behandeling van biofilm-geassocieerde infecties. Een persister is een bacterie die, onder invloed van allerlei omgevingsfactoren, in slaaptoestand gaat waardoor deze niet meer wordt herkend door antibiotica. In **hoofdstuk 7** hebben wij dit aangetoond in een experiment waarin bacteriën, ondanks langdurige behandeling met antibiotica (rifampicine gecombineerd met ciprofloxacine), bleken te kunnen overleven in een chronische biofilm. Ontwikkeling van alternatieve geneesmiddelen die persisters kunnen doden is daarom noodzakelijk om deze biofilm-geassocieerde infecties te genezen. Een dergelijk geneesmiddel zou de uitkomst van PJI en vele andere biofilm-geassocieerde infecties zoals infecties van vaatprotheses, kunstklependocarditis, fractuur-gerelateerde infecties, spondylodese infecties en geïnfecteerde pacemakers aanzienlijk kunnen verbeteren. Helaas zitten er bijna geen anti-persister geneesmiddelen in de pijplijn voor de komende jaren. In de afgelopen decennia zijn wel verschillende anti-persister behandelstrategieën ontwikkeld die in de toekomst tot klinische toepassing kunnen leiden. Antimicrobiële peptiden zijn werkzaam tegen bacteriën en lijken ook effectief te zijn tegen persisters. SAAP-148 is een antimicrobieel peptide en ontwikkeld in het LUMC. Het peptide heeft een brede werking tegen bacteriën zoals methicilline-resistente *Staphylococcus aureus* (MRSA) en Gram-negatieve bacteriën in ex vivo en in vivo wondinfecties. Dit bracht ons ertoe om preklinische onderzoeksmodellen met chronische biofilms op kunstmateriaal te ontwikkelen waarin we anti-persister geneesmiddelen konden testen. In **hoofdstuk 7** beschrijven wij hoe we een in vitro model met een rijpe biofilm ontwikkeld hebben. Het doel was om een model met een chronische biofilm te ontwikkelen die een prothese infectie zo goed mogelijk nabootst, zodat de uitkomsten uiteindelijk toegepast kunnen worden in de praktijk. Wij hebben de effectiviteit van middelen tegen persisters in biofilms geëvalueerd op meerdere kunstmaterialen: polystyreen, titanium/aluminium/niobiumschijfjes en liners van een heupprothese. De peptides (met de namen SAAP-148, acyldepsipeptide-4, LL-37 en pexiganan) bleken in staat bacteriën afkomstig uit en verblijvend in deze biofilms te doden. SAAP-148 doodde als enige peptide ook alle bacteriën uit zeven dagen oude biofilms op kunstmateriaal die een week voorbehandeling met antibiotica hadden overleefd. Dit wijst op de potentie van SAAP-148 als effectief anti-persister geneesmiddel. Dit model met een 7 dagen oude biofilm op meerdere soorten kunstmateriaal kan gebruikt worden om andere nieuwe behandelstrategieën te testen, zoals bacteriofagen, quorum sensing remmers of andere antimicrobiële peptiden. Toepassing van SAAP-148 in een oplossing op een geïnfecteerde prothese zou, als aanvullende behandeling tijdens chirurgisch debridement, een relevante klinische toepassing kunnen zijn. Dit moet verder worden onderzocht.

Conclusies

Door nauwkeurige zelf-monitoring van postoperatieve wonden na plaatsing van een gewrichtsprothese werd inzicht verkregen in het beloop van wondlekkage en het verband met acute prothese infecties. Het verzamelen van klinische gegevens over verschillende antimicrobiële behandelstrategieën heeft inzicht gegeven in de effectiviteit van verschillende behandelopties voor patiënten met een prothese infectie. Dit proefschrift toont aan dat een meer gepersonaliseerde antimicrobiële behandeling voor prothese infecties mogelijk is zonder in te leveren op de effectiviteit van die behandeling. De komende jaren zal de rol van verschillende orale behandelopties verder onderzocht worden, onder andere in de al genoemde multicenter RiCOTTA studie in Nederland. Daarnaast beschrijft dit proefschrift de rol en het belang van nieuwe anti-persister geneesmiddelen tegen biofilm-geassocieerde infecties. Wij ontwikkelden een biofilm model dat zo goed mogelijk de kliniek van een prothese infectie benadert. Op basis van de resultaten uit dit proefschrift zal onderzoek voortgezet worden, gericht op het beter begrijpen van de pathogenese van biofilms. Ook kan de effectiviteit van nieuwe geneesmiddelen tegen biofilms onderzocht worden met dit model. Dit moet leiden tot betere behandelopties voor patiënten met geïnfecteerd kunstmateriaal, waarmee uiteindelijk het doel bereikt wordt: betere zorg voor kwetsbare patiënten die geconfronteerd worden met een ernstige postoperatieve complicatie.

Nawoord

Het aantal pagina's in dit proefschrift legt het ruim af tegen de vele patiënten en medewerkers die hebben bijgedragen aan alle projecten. Samen maakten we de zorg een beetje beter. En en passant faciliteerden jullie de wetenschappelijke vorming van een clinicus. Mijn dankbaarheid voor jullie bijdrage is groot. Graag noem ik in dit slotakkoord een aantal personen die veel voor mij betekend hebben tijdens het werken aan dit proefschrift de afgelopen jaren.

Beste Mark, jouw energie, scherpte en leiderschap waren de motor achter de prachtige projecten die we uitgevoerd hebben, waarbij jij meestal mogelijkheden zag, en ik leeuwen en beren. Ik prijs me gelukkig met jou verder te kunnen bouwen aan ons nieuwe project van de RiCOTTA studie.

Wat hebben we de afgelopen jaren veel brandende vragen gesteld die opgelost moesten worden, Leo. Jij weet altijd uitmuntend tot de kern van een probleem door te dringen en de juiste vragen te stellen. Onder jouw vriendelijk leiderschap is het goed werken op de afdeling infectieziekten.

Beste Peter, ooit liep ik jouw kamer binnen na een referaat door Leo over ADEP4. Al gauw bestelden we het peptide en begon het experimenteren. Inmiddels bereikt het biofilm onderzoek onder leiding van jou en Mark de fase van dispersal. Jij leerde mij precies redeneren bij het schrijven; je vele scribbles in de kantlijnen waren altijd de moeite van het ontcijferen waard.

Beste Geert, Hetty, Merel, Anna, Sandra, Meta, Liesbeth, Natasja en Willemien, jullie collegialiteit is legendarisch en persisterend. We hebben een inspirerend klinisch topteam en ik prijs me gelukkig met jullie te mogen samenwerken.

Het vrijdagse MDO met orthopeden, microbiologen, infectiologen en traumachirurgen is de leukste bespreking van de week. Beste Robert en Rob, ik ben dankbaar voor onze hechte samenwerking van de afgelopen jaren die ons zoveel heeft opgeleverd.

Dankzij jullie, Josefien en Maxime, liep de data invoer in Castor altijd op rolletjes. Beste Brenda, zonder jouw vasthoudende vriendelijkheid naar alle centra was de Wondzorg App studie nu nog bezig. Collega's van het lab, met name Adriëtte, dank voor het helpen van een clinicus met veel wensen maar twee linker labhanden. Dankzij jullie hulp is ons mature biofilm model zonder twijfel het beste model wat er is.

Dank, paranimfen, dat jullie naast mij staan op de dag van de verdediging. Beste Martin, al oneindig lang evalueren we het leven en vieren we vriendschap, veel te weinig frequent, maar altijd goed. Floris, dank voor je zorgzame broederschap en support, op naar de volgende borrel!

Beste pa en ma, aan jullie vermogen een groot gezin draaiende te houden kunnen weinig mensen tippen. We delen de liefde voor de natuur en jullie huidige woning faciliteert die liefde ruimschoots.

Eindelijk dan papa's feest, lieve Simon, Philippa en Anna! Ik heb het even uitgesteld totdat jullie er alle drie bij mochten zijn. Maar het leven met jullie is een nog veel mooier feest.

Lieve Maaike, de laatste regels uit mijn CV zijn de belangrijkste. Wat hebben we samen bergen verzet de afgelopen jaren. Jouw vermogen om een thuis te bouwen, een plek om te zijn en te blijven, zorgt er altijd weer voor dat ik me realiseer hoe en waar ik wil leven, dicht bij jou en bij onze kinderen.

List of publications

Wouthuyzen-Bakker M, **Scheper H**, Rifampicin in periprosthetic joint infections: where do we stand and where are we headed? *Expert Review of Anti-infective Therapy* 2023, May 8;1-7.

Sanders PTJ, **Scheper H**, Van der Wal RJP, Van de Sande MJA, De Boer MGJ, Dijkstra PDS, Bus MPA, *Infected tumor prostheses of the lower extremities: causative micro-organisms, effectiveness of DAIR and risk factors for treatment failure, under review*

Van Dun S, Verheul E, Pijls BGCW, Van Prehn, J, **Scheper H**, Galli F, Nibbering PH, De Boer MGJ, *Influence of surface characteristics of implant materials on MRSA biofilm formation and effects of antimicrobial treatment*, Frontiers in Microbiology 2023, doi.org/10.3389/fmicb.2023.1145210

Gerritsen M, **Scheper H**, De Boer MGJ, Schoones J, Nelissen RGHH, Pijls BP *Outcome of debridement, antibiotics and implant retention for streptococcal hip and knee prosthetic joint infections: a systematic review and meta-analysis, under review*

Scheper H, Mahdad R, Elzer B, Löwik C, Zijlstra W, Gosens T, Van der Lugt JCT, Van der Wal RJP, Poolman RW, Somford MP, Jutte PC, Bos PK, Zwaan RE, Nelissen RGHH, Visser LG, De Boer MGJ, on behalf of the wound care app study group, *Wound leakage after arthroplasty and prediction of prosthetic joint infection: prospective data from a multicenter cohort study using a telemonitoring app*, Journal of Bone and Joint Infection, 2023, 8, 59–70

Scheper H, Van der Wal RJP, Mahdad R, Keizer, S, Delfos N, Van der Lugt JCT, Veldkamp KE, Nolte PA, Leendertse M, Gelinck LBS, Mollema PN, Schippers EF, Wattel-Louis H, Visser LG, Nelissen RGHH, De Boer MGJ *Effectiveness of Different Antimicrobial Strategies for Staphylococcal Prosthetic Joint Infection: Results From a Large Prospective Registry-Based Cohort Study.* Open Forum Infectious Diseases 2022 doi.org/10.1093/ofid/ofac474

Scheper H, De Boer MGJ. *Rifampin for staphylococcal Prosthetic Joint Infection, do we still need a RCT?* Clin Infect Dis. 2022 Apr 9;74(7):1316-1318.

Scheper H, Gerritsen, LM, Pijls, BG, Van Asten, SA, Visser, LG, De Boer, MGJ *Outcome of debridement, antibiotics and implant retention for staphylococcal hip and knee prosthetic joint infections, focused on rifampicin use: a systematic review and meta-analysis.* Open Forum Infectious Diseases 2021, 8(7): ofab298

Scheper H, Wubbolts JW, Verhagen JAM, De Visser, AW, Van der Wal RJP, Visser LG, De Boer MGJ, Nibbering PH, *SAAP-148 eradicates MRSA persisters within mature biofilm models*

simulating prosthetic joint infection, Frontiers in Microbiology 2021, doi.org/10.3389/fmicb.2021.625952

S. Schalekamp , C.P. Bleeker-Rovers, L.F.M. Beenen, H.M.E. Quarles van Ufford, H.A. Gietema, J.L. Stöger, V. Harris M.H.E. Reijers, J. Rahamat-Langendoen, D.A. Korevaar, L.P. Smits, C. Korteweg, T. van Rees Vellinga, M. Vermaat, P.M. Stassen, **H. Scheper**, R. Wijnakker , F.J. Borm, A.S.M. Dofferhoff, W.M. Prokop, *Chest CT in the Emergency Department for Diagnosis of COVID-19 Pneumonia: Dutch Experience*, Radiology 2020 Nov 17;203465. doi: 10.1148/radiol.2020203465

Scheper H, Van Asten SAG, Van der Wal RJP, De Boer MGJ, *Rifampicine bij orthopedische infecties: een historisch overzicht*, Ned Tijdschr Med Microbiol 2020;28:nr3

Scheper H, De Boer MGJ, Comment on "*Duration of rifampin therapy is a key determinant of improved outcomes in early-onset acute prosthetic joint infection due to Staphylococcus treated with a debridement, antibiotics and implant retention (DAIR): a retrospective multicenter study in France*" by Becker et al. J. Bone Joint Infect. 2020, 6, 17–18, 2020

Garrigues GE, Zmistowski B, Cooper AM, Green; Abboud J, Beasley J, Belay ES, Benito N, Cil A, Clark B, Claro R, Reinoso JC, Cvetanovich G, Del Pozo JL, Duquin T, Ekelund A, Encalada I, Entezari V, Frankle M, Garrigues GE, Grogan B, Hasan SS, Henry M, Hsu J, Itamura J, Karademir G, Kelly J, Khazzam M, King JJ, Lambert S, Ledbetter L, Levine W, Mora JM, Morrey M, Namdari S, Nelson SB, Page R, Paxton SE, Pottinger P, Romeo A, **Scheper H**, Somerson J, Tashjian R, Torrens C, Virk M, Wall KC, Willems J, Williams G, Yian E. *Proceedings from the 2018 International Consensus Meeting on Orthopedic Infections: management of periprosthetic shoulder infection*. J Shoulder Elbow Surg. 2019 Jun;28(6S):S67-S99

Welling M, Bunschoten A, Velders AH, **Scheper H**, Smits WK, Roestenberg M, Leeuwen FWB van. *Fluorescent imaging of bacterial infections and recent advances made with multimodal radiopharmaceuticals*. Clinical and Translational Imaging. 2019(7):125-38.

Scheper H, Derogee R, Mahdad R, Wal RGJ van der, Nelissen RGHH, Visser LG, De Boer, MGJ, *A mobile app for postoperative wound care after arthroplasty: ease of use and perceived usefulness*, Int J Med Inform. 2019 Sep;129:75-80

Govaert GAM, Termaat MF, Glaudemans AWJM, Geurts JAP, de Jong T, de Jong VM, Joosse P, Kooijmans H, Overbosch J, **Scheper H**, Spijkerman IJB. *Diagnosis and treatment of fracture-related infections*. Ned Tijdschr Geneeskd. 2019 Apr 11;163.

Sanders PTJ, Bus MPA, **Scheper H**, van der Wal RJP, van de Sande MAJ, Bramer JAM, Schaap GR, de Boer MGJ, Dijkstra PDS, *Multiflora and Gram-Negative Microorganisms Predominate in Infections Affecting Pelvic Endoprostheses Following Tumor Resection*, J Bone Joint Surg Am. 2019 May 1;101(9):797-803. Doi: 10.2106/JBJS.18.00836.

Carijo JH, Courtney PM, Goswami K, Groff H, Kendoff D, Matos J, Sandiford NA, **Scheper H**, Schmaltz CAS, Shubnyakov I, Tan TL, Wouthuyzen-Bakker M, *Hip and Knee Section, Pathogen Factors: Proceedings of International Consensus on Orthopedic Infections*; Journal of Arthroplasty 34 (2019) S38-386

Wijsman CA, Hanssen JLJ, **Scheper H**, Visser LG, van Lieshout L, *A case of delayed diagnosis of East-African trypanosomiasis in a Dutch traveler*, J Travel Med. 2018 Jan 1;25(1)

Uzorka JW, Delfos NM, Witte AMC, **Scheper H**, van Soolingen D, Arend SM. *Tuberculosis after a borderline QuantiFERON result during screening before infliximab*, Eur Respir J. 2018 Aug 2;52(2).

Scheper H, Van Hooven D, Van De Sande M, Van der Wal RGJ, Van Der Beek MT, Visser LG, De Boer MGJ, Nelissen RGHH, *Outcome of acute staphylococcal prosthetic joint infection treated with debridement, implant retention and antimicrobial treatment with short duration of rifampicin*, J Infect. 2018 May;76(5):498-500

Vlasveld IN, **Scheper H**, *Leptospirosis-associated facial palsy*, Netherlands Journal of Infectious Diseases 2018;13(1):23)

Kouijzer IJE, **Scheper H**, de Rooy JWJ, Bloem JL, Janssen MJR, van den Hoven L, Hosman AJF, Visser LG, Oyen WJG, Bleeker-Rovers CP, de Geus-Oei LF., *The diagnostic value of 18F-FDG-PET/CT and MRI in suspected vertebral osteomyelitis – a prospective study*. Eur J Nucl Med Mol Imaging. 2018 May;45(5):798-805

Scheper H, van Holten N, Hovens J, de Boer M., *Severe depression as a neuropsychiatric side effect induced by dolutegravir*, HIV Med. 2018 Apr;19(4):e58-e59

Vlasveld IN, **Scheper H**, Stalenhoef JS, Baas, Y, Dissel JT van, *A fatal case of metastatic squamous cell carcinoma in a patient with myositis ossificans traumatica*, Neth J Med. 2017 Jul;75(6):250-252

Kolwijck E, **Scheper H**, Beuving J, Kuijper EJ, Hoeven JG van der, Verweij PE, *Invasieve pulmonale aspergillose bij influenza*. NTvG 2015;159: A7431

Gerwen M van, **Scheper H**, Touw DJ, Nieuwkoop K van, *Life-threatening acute lung injury after gamma butyrolactone ingestion*, Neth J Med 2015 Jan 73;3:133-135

Scheper H, Lijfering WM, *Link between co-trimoxazole and sudden death in patients receiving inhibitors of renin-angiotensin system could be due to confounding*. BMJ 2014 Nov 18;349:g6899

Scheper H, van Dillen J, Blok P, Berning B. *Virilisation due to an ovarian mucinous cystadenoma*. Ned Tijdschr Geneeskd. 2007 Dec 15;151(50):2792-6.

List of participating centers

This thesis was made possible only by the collaborative efforts of nurses and doctors at all participating hospitals. I am very much indebted to everyone who contributed their time and expertise, in particular the trial participants and local research coordinators. Their valuable input and kind understanding were important in ensuring the success of the studies discussed in this thesis. This list contains all the collaborators of the regional quality registry and of the woundcare app study.

Alrijne ziekenhuis *(regional quality registry & woundcare app study)*
Drs. Rachid Mahdad, orthopedic surgeon
Dr. Masja Leendertse, medical microbiologist
Drs. Nathalie Delfos, infectious diseases physician
Dr. Machteld van der Feltz, infectious diseases physician
Drs. Menno Barnard, research coordinator
Mw. Astrid Mol, nurse orthopedic department

Haaglanden Medisch Centrum *(regional quality registry)*
Drs. Stefan Keizer, orthopedic surgeon
Dr. Luc Gelinck, infectious diseases physician
Dr. Maurine Leverstein-van Hall

Reinier-Haga Orthopedisch Centrum / Haga ziekenhuis *(regional quality registry & woundcare app study)*
Dr. Joris van der Lugt, orthopedic surgeon
Dr. Emile Schippers, infectious diseases physician
Drs. Roos Bazuin, research coordinator
Dr. Jantsje Pasma, research coordinator
Dr. Nina Matthijssen, research coordinator

Spaarne ziekenhuis *(regional quality registry)*
Prof. dr. Peter Nolte, orthopedic surgeon
Drs. Marjolein Schager, research nurse
Dr. Jayant Kalpoe, medical microbiologist
Dr. Bjorn Herpers, medical microbiologist
Drs. Hanke Wattel, internist

Leids Universitair Medisch Centrum *(regional quality registry & woundcare app study)*
Prof. dr. Rob Nelissen, orthopedic surgeon

Dr. Robert van der Wal, orthopedic surgeon
Dr. Karin Ellen Veldkamp, medical microbiologist
Mw. Brenda Elzer, research nurse & national woundcare app study coordinator
Mw. Mireille de Jong, research nurse
Dr. Enrike van der Linden, orthopedic surgeon

Medisch Centrum Leeuwarden *(woundcare app study)*
Dr. Wierd Zijlstra, orthopedic surgeon
Mw. Baukje Dijkstra, research coordinator

Onze Lieve Vrouwe Gasthuis Amsterdam *(woundcare app study)*
Prof. dr. Rudolf Poolman, orthopedic surgeon
Dr. Nienke Willigenburg, senior researcher / research coordinator

Erasmus Medisch Centrum *(woundcare app study)*
Dr. Koen Bos, orthopedic surgeon
Dr. Max Reijman, research coordinator

Universitair Medisch Centrum Groningen *(woundcare app study)*
Prof. dr. Paul Jutte, orthopedic surgeon
Dr. Claudia Löwik, resident orthopedic surgery
Dr. Martin Stevens, senior researcher

Rijnstate Ziekenhuis *(woundcare app study)*
Dr. Matthijs Somford, orthopedic surgeon
Dr. Lex de Jong, senior research coordinator

Elizabeth Tweestedenziekenhuis *(woundcare app study)*
Dr. Taco Gosens, orthopedic surgeon
Mw. Liesbeth Paans, physician assistent
Drs. Wouter Bekkers, orthopaedic surgeon in training
Nij Smellinghe Ziekenhuis *(woundcare app study)*
Drs. Carolien Kooijman, orthopaedic surgeon
Mw. Hendrike Hoogeboom, physician assistant

Park Medisch Centrum *(woundcare app study)*
Drs. Hieron Maree, financial director
Mw. Chris Meier, research student

Curriculum Vitae

Henk werd geboren op 25 maart 1980 in Oostflakkee. Hij groeide op in Hendrik-Ido-Ambacht en Rotterdam-Zuid met zijn zussen Margriet, Lenneke, Liesbeth, Willie, Alie, Maaike, Eline en broers Floris, Jaap, Adriaan en Johannes. Na het behalen van zijn VWO diploma aan de Guido de Brès scholengemeenschap in Rotterdam startte hij met de studie psychologie aan de Universiteit Leiden.

Na zijn propedeuse psychologie ging hij geneeskunde studeren in Leiden. Zijn coschappen liep hij in Leiderdorp, Leiden en Den Haag. Na zijn artsexamen deed hij klinische ervaring op als ANIOS in het Alrijne ziekenhuis in Leiderdorp. Vervolgens werkte hij een jaar in Christchurch Hospital, Nieuw Zeeland, als senior house officer op de afdelingen interne geneeskunde en neurologie. Bij zijn terugkomst in Nederland startte hij in 2009 met de opleiding tot internist (opleiders prof. dr. Hans Romijn, prof. dr. Jaap van Dissel). Hij deed zijn vooropleiding in het Haga Ziekenhuis (opleider dr. Maarten van Aken) waar zijn passie voor infectieziekten ontstond. Hij vervolgde de opleiding daarom met het aandachtsgebied infectieziekten (opleider prof. dr. Leo Visser). Tijdens zijn periode als fellow raakte hij steeds meer geïnteresseerd in de behandeling van patiënten met complexe bot- en gewrichtsinfecties en infecties met betrokkenheid van kunstmateriaal.

Sinds 2016 werkt hij als internist-infectioloog in het LUMC en begon hij, naast zijn poliklinische en klinische werkzaamheden, aan een promotietraject onder leiding van prof. dr. Mark de Boer en prof. dr. Leo Visser. Hij maakte deel uit van de Nederlandse richtlijncommissie voor fractuur-gerelateerde infecties, de internationale consensus meeting prosthetic joint infections in Philadelphia (2018) en participeert in de huidige SWAB richtlijncommissie voor geïnfecteerde gewrichtsprotheses. Samen met Maaike Snoep woont hij in Voorburg. Ze hebben drie kinderen: Simon, Philippa en Anna.

Stellingen bij het proefschrift

Prosthetic Joint Infections: new diagnostic and therapeutic strategies

1. De superioriteit van rifampicine ten opzichte van andere antibiotica voor de behandeling van gewrichtsprothese infecties veroorzaakt door stafylokokken is niet aangetoond in klinische studies *(dit proefschrift)*.
2. Het vergelijken van één specifieke behandeling (rifampicine) met alle overige behandelingen in een gezamenlijke 'niet-rifampicine groep' leidt tot overschatting van het effect in de specifieke behandelgroep *(dit proefschrift)*.
3. Wondlekkage na plaatsing van een gewrichtsprothese heeft een lage positief voorspellende waarde voor het optreden van een acute prothese infectie *(dit proefschrift)*.
4. Met antimicrobiële peptides, zoals SAAP-148, kunnen persisters in mature biofilms geëradiceerd worden *(dit proefschrift)*.
5. De veel toegepaste praktijk van het uitstellen van de behandeling met rifampicine na debridement van een geïnfecteerde prothese door stafylokokken is niet logisch.
6. Bij verdenking op geïnfecteerd kunstmateriaal dienen positieve kweken met een *Cutibacterium acnes* even serieus genomen te worden als kweken met een *Staphylococcus aureus*.
7. Het verschil tussen bacteriostatische en bactericide antibiotica is alleen in uitzonderlijke klinische situaties relevant.
8. Het voorschrijven van levenslange suppressieve antibiotische therapie bij chronisch geïnfecteerd kunstmateriaal is niet evidence-based.
9. Hoe sterker de wens dat een behandeling effectief is hoe groter het risico dat bij de wetenschappelijke onderbouwing voor die behandeling onvoldoende gecontroleerd wordt voor potentiële confounders (naar Paul Sax, blog *HIV and ID observations*)
10. Vogels kijken is de mooiste variant van observationeel onderzoek waarbij zorgvuldige observatie en geduld een sleutel zijn tot succes en gelukservaringen. (naar Tish Warren, *Prayer in the Night*, p.58)
11. Hardlopen is beter voor de geest dan voor het lichaam.