Acta Biomaterialia xxx (xxxx) xxx

Contents lists available at ScienceDirect

Acta Biomaterialia

journal homepage: www.elsevier.com/locate/actbio

Review article

Megaprosthesis anti-bacterial coatings: A comprehensive translational review

Johnathan R Lex[a,1], Robert Koucheki[b,c,1], Nikolaos A Stavropoulos[d,*], Joseph Di Michele[a], Jay S Toor[a], Kim Tsoi[a,c,e], Peter C Ferguson[a,e], Robert E Turcotte[f], Panayiotis J Papagelopoulos[g]

[a] Division of Orthopaedic Surgery, Department of Surgery, University of Toronto, Toronto, Canada
[b] Temerty Faculty of Medicine, University of Toronto, Toronto, Canada
[c] Institute of Biomedical Engineering, University of Toronto, Toronto, Canada
[d] Department of Orthopaedic Surgery, General Hospital of Karpenisi, Karpenisi, Greece
[e] University Musculoskeletal Oncology Unit, Mount Sinai Hospital, Toronto, Canada
[f] Division of Orthopedic Surgery, McGill University Health Centre, Montreal, Canada
[g] 1st Department of Orthopaedic Surgery, School of Medicine, National and Kapodistrian University of Athens, Greece

ARTICLE INFO

Article history:
Received 31 July 2021
Revised 29 November 2021
Accepted 30 November 2021
Available online xxx

Keywords:
Periprosthetic joint infection
Implant coating
Prevention
Orthopaedic oncology
Joint replacement
Bacterial biofilm
Orthopaedic surgery

ABSTRACT

Periprosthetic joint infections (PJI) are catastrophic complications for patients with implanted megaprostheses and pose significant challenges in the management of orthopaedic oncology patients. Despite various preventative strategies, with the increasing rate of implanted orthopaedic prostheses, the number of PJIs may be increasing. PJIs are associated with a high rate of amputation. Therefore, novel strategies to combat bacterial colonization and biofilm formation are required. A promising strategy is the utilization of anti-bacterial coatings on megaprosthetic implants. In this translational review, a brief overview of the mechanism of bacterial colonization of implants and biofilm formation will be provided, followed by a discussion and classification of major anti-bacterial coatings currently in use and development. In addition, current in vitro outcomes, clinical significance, economic importance, evolutionary perspectives, and future directions of anti-bacterial coatings will also be discussed. Megaprosthetic anti-bacterial coating strategies will help reduce infection rates following the implantation of megaprostheses and would positively impact sarcoma care.

Statement of significance

This review highlights the clinical challenges and a multitude of potential solutions to combating periprosthetic join infections in megaprotheses using anti-bacterial coatings. Reducing infection rates following the implantation of megaprostheses would have a major impact on sarcoma care and major trauma surgeries that require reconstruction of large skeletal defects.

1. Background

Due to the advances in systemic therapy, imaging, surgical technique and endoprosthesis technology, limb-salvage surgery (LSS), is considered the gold-standard treatment strategy following large bone resection for benign and malignant tumours [1]. Over the last two decades, the rate of LSS has been increasing, while overall survival rates and indication for secondary amputation following LSS resulting from postoperative complications have decreased [2]. In addition, compared to extremity amputation, LSS offers superior limb function and quality of life, without impacting overall survival [3–7]. However, the incidence of post-operative infection remains high and presents several unique management challenges [8]. Rates of periprosthetic joint infection (PJI) in non-oncologic arthroplasty in Western settings, has been estimated to be around 1.2–2.2%, [9] while rates of PJI in oncology procedures have been estimated to range from 7%-28% [10,11]. The higher prevalence of infections in LSS is likely multifactorial. Contributing factors include local and systemic immunodeficiency resulting from the primary cancer and immunosuppressive therapeutics

* Corresponding author at: 2 P.Bakoyianni, 36100, Karpenisi, Greece.
E-mail address: stavropoulosna@gmail.com (N.A. Stavropoulos).
[1] Joint first author.

https://doi.org/10.1016/j.actbio.2021.11.045
1742-7061/

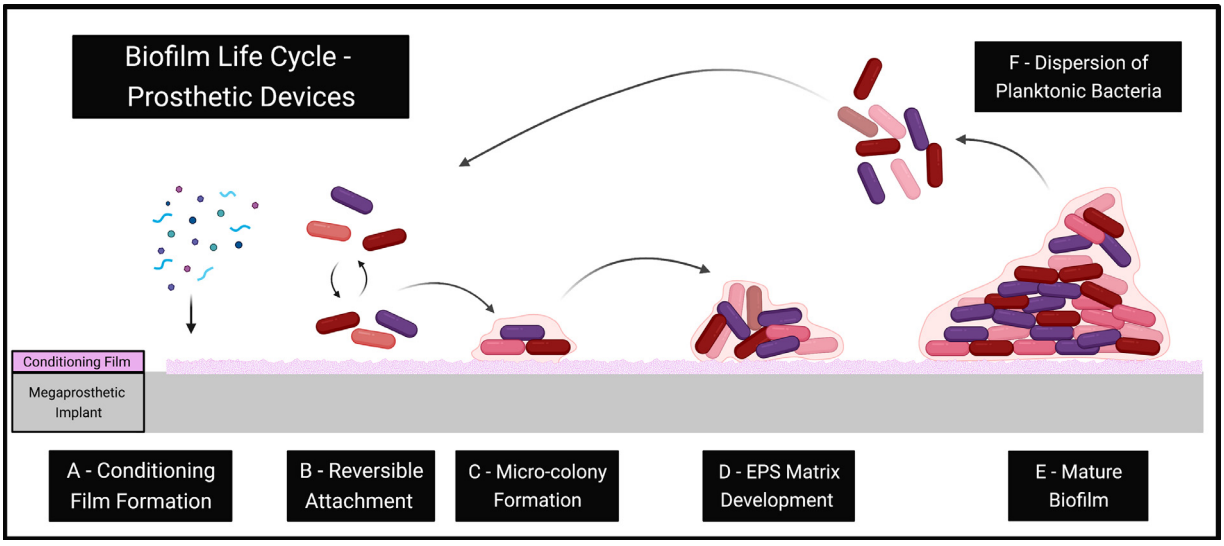

Fig. 1. Biofilm life cycle.

such as chemotherapy and radiotherapy, longer surgical durations, large surgical incisions, soft-tissue dead space, and use of large metallic implants, also known as megaprostheses [10,12,13]. Other risk factors for development of PJI include patient factors, such as high BMI and diabetes, and certain anatomic locations, such as pelvic and tibial prostheses [8,11,13,14]. Infection of megaprostheses are catastrophic for patients, often requiring lengthy treatment courses, multiple revision surgeries, and are associated with high failure rates, leading to amputation in 30% to 40% of cases [8,15]. PJIs also pose a significant economic burden on the health care system [16]. Current preventative strategies include pre-operative skin cleansing and MRSA screening, prophylactic pre- and post-operative antibiotics, sterile field prepping and draping, laminar airflow operating theatres [17,18]. A potential strategy to address the high rates of PJIs and megaprosthetic infections is the introduction of anti-microbial coatings on the surface of implants [19]. In this review we will first provide an overview of important considerations related to biofilm formation and PJIs, provide a summary of the major classes of antimicrobial coatings, and describe the current clinical applications of these coatings.

2. Bacterial colonization of implants and biofilm formation

For bacterial colonies to become established on implants, a threshold infectious dose of bacteria is required [20]. This threshold dose is dependent upon the bacterial virulence, host immune response, and amount of necrotic tissue available for bacterial colonization [15]. Once implants are introduced *in vivo*, proteins and glycoproteins (such as complement, albumin, fibronectin, fibrinogen, laminin, collagen, and von Willebrand factor) followed secondarily by polysaccharides, adsorb to the surface of the implant, creating a layer called the conditioning film (Fig. 1-A) [21–24]. At this stage, there is a competition between host eukaryotic cells and planktonic bacteria to colonize the implant areas covered with the conditioning film. Adhesion of planktonic bacteria to the implant surface is influenced by the implant material, implant surface topography, local temperature, pressure, and bacterial cell wall properties [22,25,26]. At this stage antibiotics are effective at inhibiting and decreasing the number of planktonic bacteria to prevent PJIs, however, once bacteria deposit upon the conditioning film and create a biofilm, they become highly resistant to antibiotic therapy due to protection provided by the biofilm architecture, and may require implant removal. At time of revision surgery for infection, surgeons are unable to reliably remove the entire biofilm. This often necessitates physical removal of the implant, leading to more bone loss and patient morbidity [27].

Biofilm formation occurs in different stages. Initially, planktonic bacteria start to reversibly adhere to the conditioning film through Van der Waals forces, hydrophobic and electrostatic interactions, and finally protein adhesions (Fig. 1-B) [21]. Bacterial appendages such as flagella, pili, fimbriae, and glycocalyx help the bacteria adhere firmly to the surface [28–31]. In the second stage, the adhered bacteria start to form microcolonies and produce extracellular polymeric substances (EPS) matrix (Fig. 1-C). Initially the EPS mostly consists of extracellular DNA (eDNA), however, in later stages polysaccharides and structural proteins become more prevalent [32]. Through quorum sensing and cell-cell interactions, bacteria interact with each other and further proliferate and enhance the EPS matrix leading to the formation of the mature biofilm (Fig. 1-D/E) [33,34]. Upon maturation of the biofilm, the main body starts to release planktonic bacteria into the microenvironment, further continuing the cycle and expanding the biofilm coverage (Fig. 1-F) [33,34]. Within different regions of the biofilm there exist heterogenous bacterial sub-populations, mimicking a multicellular organism, with each sub-population fulfilling a different role in the survival, maintenance, and growth of the entire bacterial biofilm community [35]. Various types of bacterial sub-populations have been recognized, including metabolically dormant sub-populations residing deep within anoxic regions of the biofilm resistant to antibiotics, structural sub-populations, and shared resource producing sub-populations [35]. Each of these subpopulation within the biofilm contributes to the various strategies that confers antibacterial resistance to the bacterial population. Some of these strategies include production of protective capsules or glycocalyx, production of anti-biotic degrading/detoxifying enzymes and efflux pumps, quorum signalling, and heterogeneity in metabolism, growth rate, and genetic adaptations within the subpopulations in response to antibiotic stress allowing for the survival of persister cells [36,37].

3. Classification of periprosthetic joint infection

Diagnosis of PJI remains difficult, with current strategies utilizing a combination of serum and synovial biochemical and microbiological parameters [38,39,40]. There are different classifications of PJI in the literature [41–43]. Generally, PJI can be classified as follows: [42]

Table 1
Desirable properties of anti-bacterial coating of megaprostheses.

Coating Properties
1. Biocompatibility and absence of local and systemic toxicity
2. Efficacious anti-bacterial activity
3. Durable anti-bacterial activity
4. Prevention of any compromises in fixation efficacy of the implant
5. Does not compromise implant mechanical stress and strain resistance properties
6. Lack of detrimental effects on bone healing and tissue integration
7. Lack of pro-tumorigenic effects
8. Cost-effectiveness

- **Early Infections**: infections that develop less than 3 months post-operatively.
- **Delayed Infections**: infections occurring between 3 to 24 months post-operatively.
- **Late Infections**: infections occurring more than 24 months post-operatively.

Early infections are typically caused by virulent organisms, such as *Staphylococcus aureus* and some gram-negative bacilli. These typically present acutely with erythrocyte sedimentation rate (ESR) elevation, joint pain, swelling, redness, and fever [11,42]. However, delayed infections are typically caused by less virulent species such as coagulase negative *Staphylococci* or *Cutibacterium acnes*, manifesting in a subtle manner with persistent bone pain and radiographic signs of implant loosening [42]. Delayed infections are more difficult to detect as they can mimic aseptic failure [42]. Early and delayed PJI infections are typically caused by bacterial seeding of the implant intra-operatively [41–43]. However, late infections occurring after 24 months are commonly caused by hematogenous spread of bacteria from skin, respiratory tract, dental, or genitourinary infections [16,42,43].

4. Summary of major anti-microbial coatings

In addition to traditional practices such as creating a sterile operating room environment and use of local and systemic antibiotic therapy, numerous novel strategies have been proposed to address PJIs. These include use of bacteriophages targeting specific bacteria, use of pre-operative vaccines targeted at common bacterial culprits, and implant surface modifications [26,44–46]. This review will focus on the surface modification strategies that prevent bacterial adhesion, colonization, and proliferation. As a result of high rates of PJIs in LSS surgery, bacteria-resistant megaprostheses would be a highly valuable addition to the armamentarium of orthopaedic surgeons. Some desirable features of anti-bacterial coatings of megaprostheses are demonstrated in Table 1. A critical consideration in the design of orthopaedic antibacterial coatings is to ensure the implant inhibits bacterial adhesion while not impairing osseointegration or osteogenesis [47]. However it should also be noted that only certain parts of the megaprosthetic implant usually needs to promote osseointegration, while for the rest of the implant anti-bacterial properties can dominate.

Broadly, implant coatings can be classified into three major categories; (A) passive anti-adhesive (anti-fouling) modifications that rely on repulsion of microbes, (B) active antimicrobial approaches that attempt to kill the microorganism, and (C) approaches that affect biofilm architecture, which focus on reducing biofilm virulence factors (Fig. 2) [48,49].

4.1. Passive anti-adhesive coating / anti-fouling

There are various passive strategies to prevent adhesion of bacterial species to the implant surfaces. Below we discuss these major passive strategies that have been studied:

4.1.1. Type of implant alloy

The two major types of megaprostheic implant alloys in clinical use are cobalt-chromium (Co-Cr) and titanium [50]. Both animal studies and clinical data have demonstrated that Co-Cr alloys have higher rates of PJIs compared to titanium alloys [50–52]. A potential explanation is the observation is that Co-Cr alloys lead to an inhibition of the local innate immune response, including a deficiency in the local monocyte-macrophage system and impaired respiratory burst of neutrophils [50–52]. Another potential contributing factor is that Co-Cr alloys have lower bio-compatibility compared to titanium alloys, leading to impaired tissue integration and opportunity for being seeded by planktonic bacteria [50]. Furthermore, titanium implants have been shown to have lower rates of biofilm formation, compared to Co-Cr implants [53].

Surprisingly, a recent study on spinal implants compared Co-Cr alloy with titanium implants and showed that Co-Cr implants suppressed *S. aureus* and *Propionibacterium acnes* proliferation and reduced micro-organism survival compared to titanium implants, in both in vitro and in vivo pre-clinical studies [54]. Therefore, further studies are required to compare these two alloy types in prevention of PJIs.

4.1.2. Polymer coatings

Hydrophilic polymeric brushes – i.e. highly hydrated polymers, can reduce protein adsorption, conditioning layer formation, and bacterial adhesion to the implant surface [47]. Polyethylene glycol (PEG) and polyethylene oxide (PEO) are frequently used for this purpose. For example, titanium coated with PEGylated titanium-binding peptides (TBPs) has been shown to impair fibronectin adsorption and *S. aureus* colonization [55,56]. Another highly hydrophilic polymer coating for titanium implants is poly(methacrylic acid) (P(MAA)). Adhesion of *S. epidermidis* and *S. aureus* to P(MAA)-modified titanium is about 3-4 times less than pure titanium. However, a major limitation of hydrophilic polymeric brushes is that they prevent the attachment of osteoblasts [57,58]. This raises caution for the utilization of these coatings on orthopaedic implants.

A promising biocompatible polymeric coating that selectively impairs bacterial adhesion while enhancing osteoblast function, is chitosan [59]. Chitosan has demonstrated anti-bacterial activity against both gram-positive and gram-negative organisms [60]. Due to its cationic charge, it interacts with the negatively charged bacterial cell wall leading to bacterial death. It has also been shown to impair bacterial DNA and RNA synthesis [61]. Titanium coated with chitosan and polyanionic hyaluronic acid has been shown to prevent bacterial adhesion and enhance osteoblast proliferation in vitro (Fig. 3) [59]. In addition, conjugating a RGD peptide (Arg-Gly-Asp) motif to the chitosan coating can further enhance osteoblast binding, without affecting bacterial adhesion [62]. A molybdenum diselenide chitosan titanium implant has been shown to decrease *Streptococcus mutans* infection in dental implants [63]. Another example is a multi-layer biopolymer of chitosan and pectin nanocomposite with silver nanoparticles, that has demonstrated anti *S. aureus* proliferation and adhesion activity.

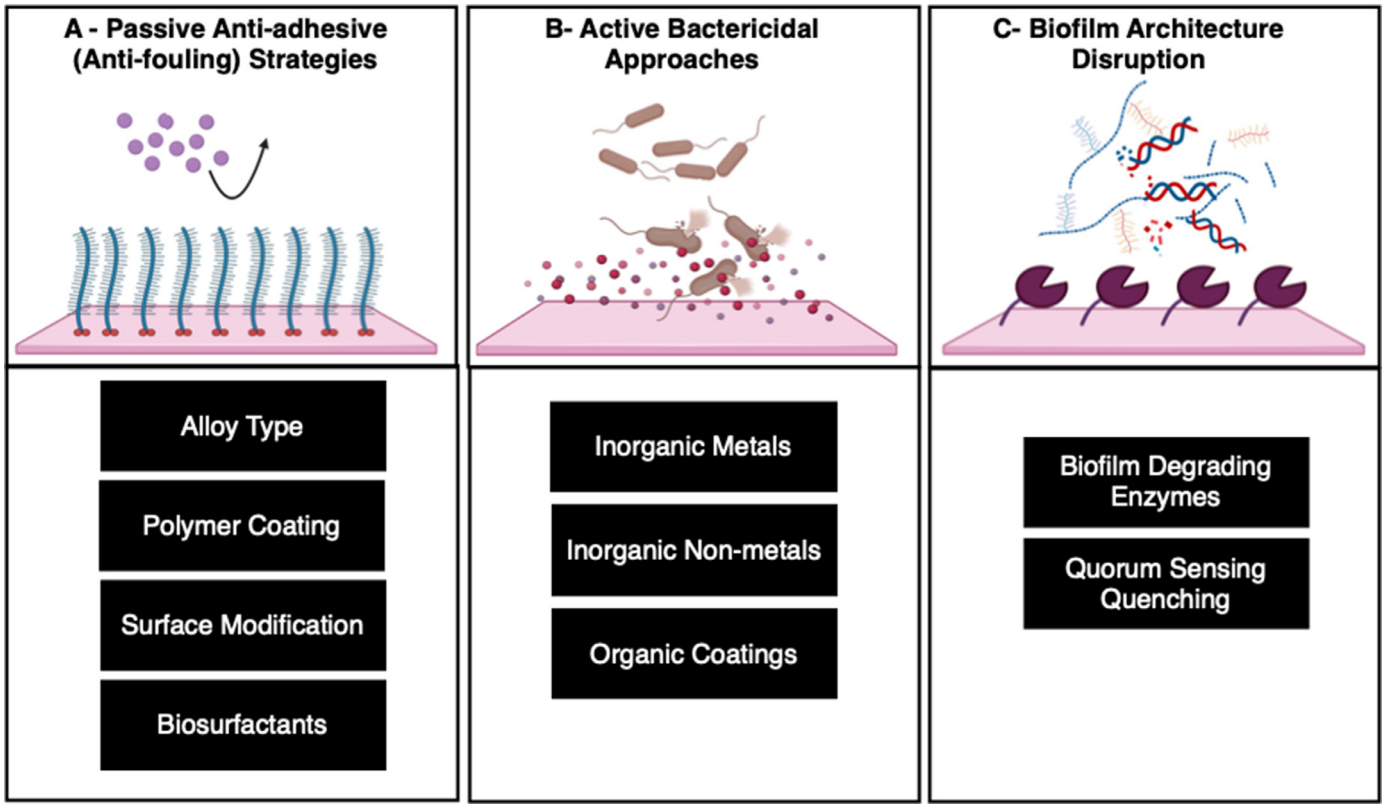

Fig. 2. Major categories of anti-microbial coatings.

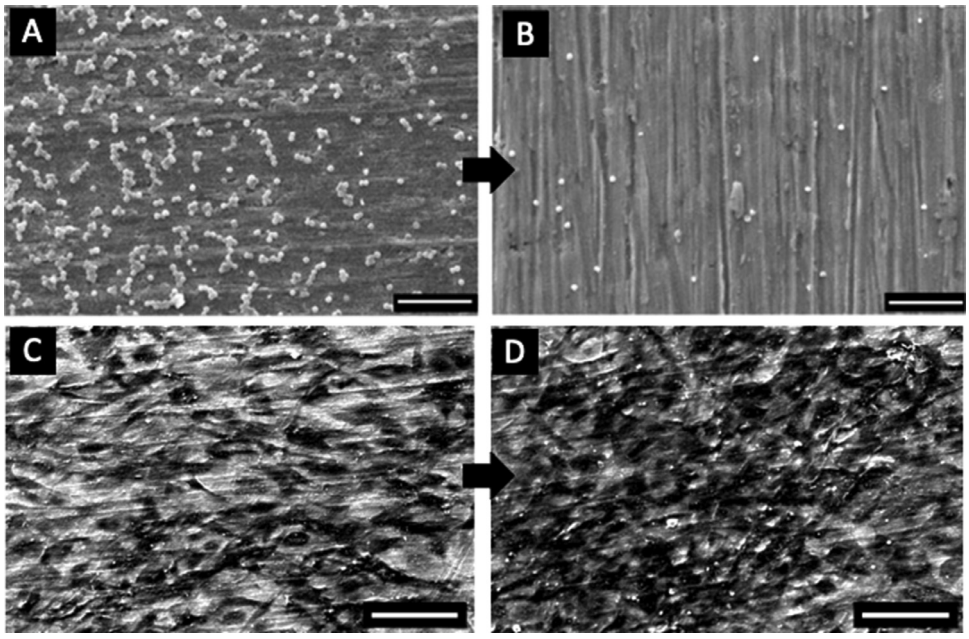

Fig. 3. A) Pure titanium in *S. aureus* suspension B) Titanium coated with five layers of chitosan and hyaluronic acid in *S. aureus* suspension. C) Osteoblast proliferation on pristine titanium D) Osteoblast proliferation on titanium coated with five layers of chitosan and hyaluronic acid. Figures A/B scale bar = 10 µm; figures C/D scale bar = 100 µm. Reproduced with permission [59].

Another class of polymeric coatings are the Defensive Antibacterial Coating (DAC®), these hydrogel coatings are made of covalently linked hyaluronan and poly-D,L-lactide, and can protect implant material as an effective barrier. [64,65]. These hydrogels can be combined with antibiotics at the time of implantation to increase protection against bacterial colonization [64].

4.1.3. Modification of implant physiochemical surface properties

By altering the surface characteristics such as roughness, hydrophobicity, and surface energy, bacterial adhesion may be inhibited [49]. For example, it has been demonstrated that ultraviolet (UV) light irradiation of titanium dioxide, increases its spontaneous wettability, resulting in inhibition of bacterial adhesion while preserving surface osteogenesis on titanium alloy implants [49,66,67].

A fascinating modification of surface properties is patterned surface topography changes. Various natural surfaces such as shark and worm skin, lotus and taro leaves, and butterfly wings have intrinsic anti-adhesive properties [68,69]. By mimicking these nano and micro-structures, implant surfaces can be fabricated that prevent bacterial adhesion. [68,69]. These designs can be achieved by lithography or hydrothermal treatments to create nanostructured bioinspired geometries [70]. Superhydrophobic anti-adhesive surfaces can be obtained by combining patterned micro/nano scaled surface topographies with hydrophobic chemical moieties [48]. For example, hydrophobic fluoroalkyls have been attached to nanostructured TiO2 surfaces, creating a superhydrophobic implant surface that reduced *S. aureus* adhesion, however this may also come at the cost of reduced osteoblast adhesion and osseointegration [71].

Another example of physiochemical modification strategies include altering the crystalline structure of the implant oxide layer; it has been previously demonstrated that modifying the crystalline anatase titanium oxide layer significantly reduces bacterial attachment. [72]. Most of these strategies are in pre-clinical stages.

4.1.4. Biosurfactants

Biosurfactants are microbial amphiphilic polymers that exhibit emulsifying activity [73]. Biosurfactants have been extracted from various bacterial species to inhibit biofilm formation, for example *Bacillus subtilis* and *Bacillus licheniformis* produce lipopeptide biosurfactants capable of reducing *Escherichia coli* and *S. aureus* biofilm formation on polystyrene surfaces by 97% and 90%, respectively [74]. Rhamnolipid biosurfactant can be physically adsorbed on titanium discs. This has been tested on several different commercially available dental implant surfaces and was found to be effective in reducing Staphylococcal biofilm formation [75]. Biosurfactants are biodegradable, reduce toxicity, are biocompatible, and effective at a wide range of temperatures and different environmental pHs. These agents are at a pre-clinical stage [73].

4.2. Active anti-microbial strategies

A common approach to inhibit biofilm formation is modifying surfaces with antibacterial agents, which are released over time. These agents can be inorganic such as metal ions, or organic coatings such as antibiotic impregnated implants.

4.2.1. In-organic coatings–transition metals

Antimicrobial toxicity of transition metals, is a result of four major chemical processes, including coordination chemistry, hard–soft acid base (HSAB) theory, reduction potential, and speciation [76]. Through coordination bonding, metal ions bond to donor atoms such oxygen, nitrogen, or sulfur. [76]. HSAB predicts metal reactivity, for example, soft acids and borderline acids such as silver, copper, and zinc associated with soft bases such as sulphhydryl groups (-SH) [77]. Fig. 4 displays some transition metal affinities for different protein moieties.

Another important property of metal ions is their ability to partake in redox reactions [76]. Furthermore, metal ions exist in different ionic states depending on the environmental conditions e.g. Cu^{+} and Cu^{2+} (speciation) [76]. These various properties allow metal ions to interfere with bacterial machinery and ultimately lead to toxicity.

Silver, zinc, and copper are common transition metals used in prosthetic surface coatings and will be discussed further.

4.2.1.1. ***Silver***. Silver coating has attracted a great deal of interest [78]. Silver has broad, long-lasting antimicrobial activity against bacteria, fungi, protozoa, and even certain viruses [79]. Active silver (Ag^{+}) can directly damage cell membranes leading to membrane perforations. Perforations lead to loss of nutrients and cellular components [80]. Silver also disrupts the electron transport chain due to its affinity for the sulfhydryl and thiol groups (SH), impairing enzyme functions, which increases reactive oxygen species (ROS) generation (Fig. 5) [81]. Additionally, silver and other transition metals can displace bacterial innate catalytic and structural metals further impairing cellular function [76]. Silver may also limit transcription and translation by binding to nucleosides, as well as causing DNA breaks [76,82]. Silver coating can be divided into two major types; ionic silver such as silver nitrates or silver chlorides in solution, or colloidal silver nanoparticles [83–85]. Silver nanoparticles have a higher antimicrobial efficacy [83,84].

Silver surface modification and coating of orthopaedic implants can be done using various strategies including anodization, galvanic electroplating, magnetron sputtering, and silanization (Table 2) [86].

Silver has also been loaded into PLGA coatings for titanium implants leading to a reduction of survival of methicillin-resistant *S. aureus* (MRSA) and *Pseudomonas aeruginosa* in a rabbit orthopedic implant infection model, while displaying osteo-inductive activity [92]. In mouse models silver coating of titanium-aluminum-niobium implants prevented perioperative infections and prevented infections after a challenge with 2×10^{6} CFU of *S. epidermidis* [93]. The same implant combined with systemic daptomycin prophylaxis was also able to prevent 100% of *S. aureus* infections [93]. Compared to other antimicrobial metals, silver has the highest antibacterial activity and the highest associated eukaryotic cytotoxicity [94]. Of note, there have been reports of bacterial resistance to silver in clinical isolates; [95]. however, silver coated implant-related bacterial resistance has yet to be reported. As silver coated implants are being utilized more in clinical settings the likelihood of evolution of silver resistance increases. *See evolutionary lens section.*

4.2.1.2. Zinc. In vitro studies have suggested that titanium-zinc coatings may be a suitable candidate for orthopedic and dental implants, as they have strong antibacterial activity, and biocompatibility [96–98]. In addition, zinc-implanted titanium has been shown to have osteogenic activity [97].

4.2.1.3. ***Copper***. Copper also has antimicrobial activity [99]. Similar to silver, copper disrupts the bacterial membrane, leading to cell rupture and loss of membrane potential [99]. Additionally, copper induces the production of ROS leading to further cellular damage [99]. In an in vitro study of orthopaedic implants made of titanium-copper-nitride coatings, *S. epidermidis* growth was completely inhibited while osteoblast colonization was favoured [100]. An important consideration is toxicity of antimicrobial metals to the eukaryotic cells [101]. It has been suggested that copper-titanium compounds may be superior to other antimicrobial metals such as silver, zinc, aluminum, and cobalt, as they have antimicrobial activity and a relatively lower degree of toxicity towards human cells [102,103].

An in vitro study has demonstrated that combinatorial therapy with silver, zinc, copper, and other transition-metal, has shown synergistic antimicrobial function compared to each individual metal alone. In addition, the combinatorial application may have decreased cytotoxicity due to lowered overall minimum inhibitory concentration for each metal by the proper formulation of synergistic metals. This leads to effective therapies that are not concentrated enough to damage eukaryotic cells while synergistically working together to inhibit bacterial proliferation [104,105].

4.2.2. In-organic coatings–non-metals

4.2.2.1. Iodine. A promising field is iodine coating of titanium alloys [106]. Iodine-supported titanium implants have effective anti-

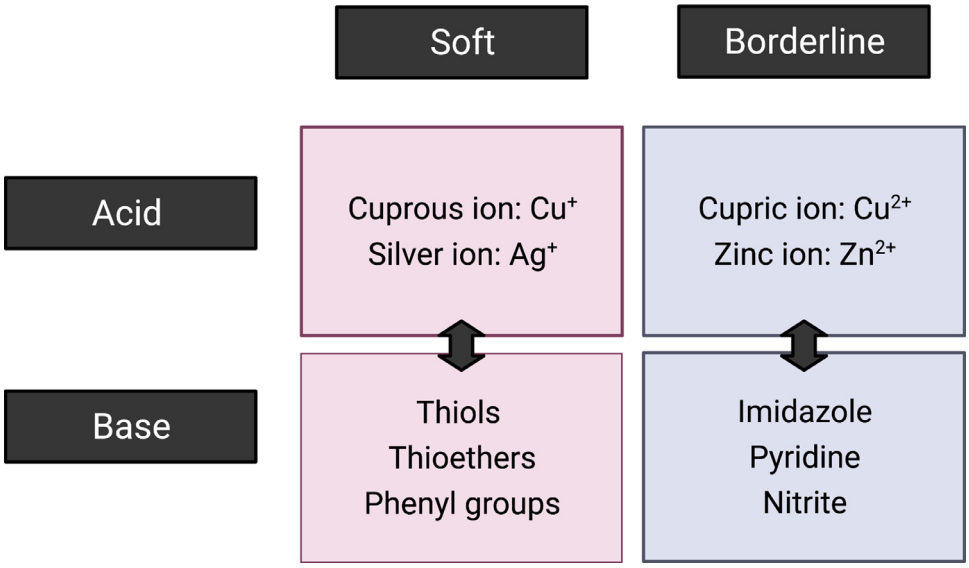

Fig. 4. Hard–soft acid base (HSAB) theory predicts the selectivity of transition metal ions for biological donor ligands – Soft acids such as Cu^{+} and Ag^{+} have affinity for soft bases such as thiol containing groups and borderline acids such as Zn^{3+} and Cu^{2+} have affinity for borderline bases such as imidazole moieties.

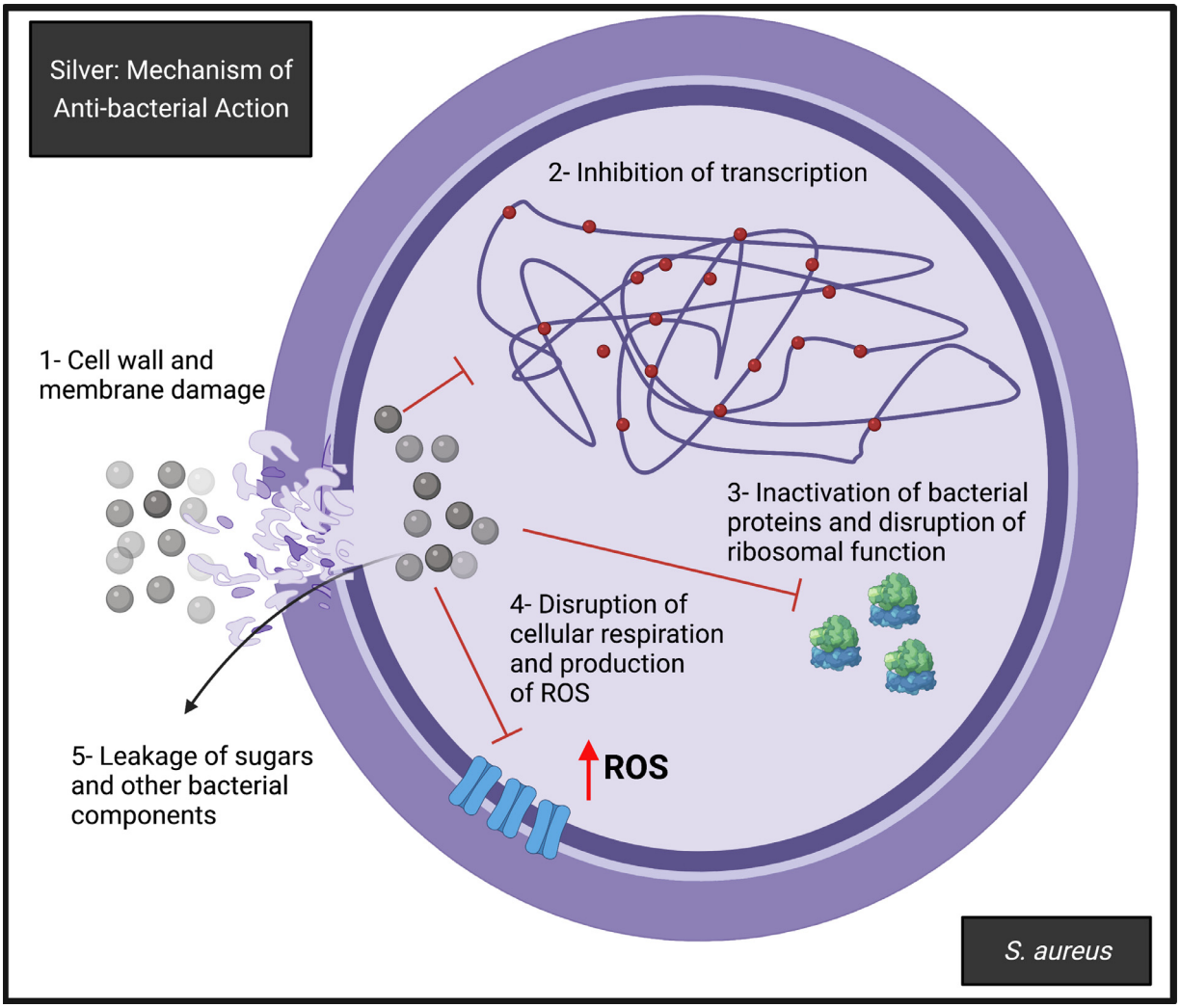

Fig. 5. Mechanism of action of silver's anti-bacterial activity – shown in the context of *S. aureus.*

Table 2
Techniques for coating prostheses with silver.

Strategies	Description
Anodization	Adsorption of aqueous ionic silver onto titanium implants using high voltages, leading to the formation of an oxide layer. This strategy leads to formation of pits on the titanium implant surface that serve as "silver release reservoirs" [86]. This method is a surface modification and not a true coating strategy because majority of the silver is gradually released in tissue leaving a behind pure titanium implant [86].
Galvanic Electroplating	In this strategy high quantities of high-purity silver are deposited on the megaprostheses titanium alloy by galvanization [87]. These coatings can be pH sensitive and increase silver ion release in response to local acidosis caused by bacteria [87]. Additionally, increase in silver release caused by low pH will be visually noticeable as a colour change in future revision surgeries [87].
Magnetron sputtering	This is a physical vapor deposition (PVD) coating strategy that uses a strong magnetic field and vaporized silver to bombard the prosthesis surface in a vacuum [86,88,89].
Nanoparticle Silanization	Silanization is a method that can be used to covalently coat titanium, hydroxyapatite, and other metal surfaces with nanoparticles using silicon [90,91].

microbial properties against *S. aureus, P. aeruginosa*, MRSA, and *Candida Albicans* [107]. Iodine-supported titanium implants have been shown to prevent and treat infection in patient with compromised immune systems or active PJIs, without any clinically detectable cytotoxic or adverse effects in over two-hundred patients with an average of 18 months of follow up [108]. Of note, even after one year, the amount of iodine on external fixation pins remained about 20-30% of the initial volume, indicating long-term stability of the coating [108].

4.2.2.2. ***Selenium.*** Another area of research is the use of selenium nanoparticles as anti-infective implant coatings for orthopedic implants. It is hypothesized that selenium kills bacteria by reacting with ROS [109]. In vitro and in vivo studies have demonstrated selenium nanoparticles are effective in preventing MRSA and *S. epidermidis* biofilm formation [110]. Selenium does not inhibit osteoblast function while inhibiting biofilm formation [111].

4.2.2.3. Nitric oxide (NO). NO was first shown to disrupt biofilm formation by modulating c-di-GMP levels in *P. aeruginosa* [112]. An in vitro study has shown NO-releasing titanium coatings on orthopaedic implants have been able to achieve maximum antimicrobial efficacy with minimum cytotoxicity to human primary osteoblasts [113]. These implants are currently in preclinical testing.

4.2.2.4. Antiseptics. Antiseptic coatings are another potential bactericidal coating strategy. and chloroxylenol coatings have been demonstrated to reduced external fixator pin tract infections, in a goat model [114]. In a rat model, chlorhexidine-coated implants reduced the overall bacterial colonisation, reduced osteolysis and increased the radiographic union. However, when the chlorhexidine-coated implant was introduced into a sterile wound, non-union increased [115]. This may be attributed to chlorohexidine inducing a local inflammatory response leading to decreased osteoconductive effects, contributing to non-union [116]. More studies are required to elucidate the biologic effects of antiseptic coatings in orthopaedic implants.

4.2.3. Organic coatings

4.2.3.1. Antibiotic coated prostheses. Antibiotics can be adsorbed onto the titanium surface of the prosthesis or be impregnated in bone cements [48]. To coat prostheses with antibiotics, biodegradable materials are used to coat the surface of titanium. For example poly-lactic-co-glycolic acid (PLGA) and poly(D,l-lactide) (PDLLA) polymers can be used to coat the implant surface [117–119]. More recently, a degradable PLGA gentamicin-loaded coating for hydroxyapatite (HA)-coated cementless hip prostheses was developed and shown to significantly reduce rates of infection compared to HA-coated implants without gentamicin in rabbit models [117]. Bone cements are also commonly impregnated with various antibiotics, most commonly gentamicin, tobramycin, or a combination of gentamicin and other antibiotics such as vancomycin [48,120–122].

4.2.3.2. Antimicrobial peptides. Antimicrobial peptides are effector proteins produced by a wide range of organisms, from prokaryotes to eukaryotic. Another strategy to counteract bacterial colonization is using antimicrobial peptide coatings. An example is a titanium implant coated with GL13K antimicrobial peptide, which is a protein derived from the parotid gland that has bactericidal and bacteriostatic properties [123]. Another study used a layer-by-layer assembly of polymer thin films with ponericin G1, an antimicrobial peptide with strong activity against *S. aureus*, and showed inhibition of adhesion and biofilm formation [124]. Antimicrobial peptides are at a pre-clinical stage.

4.2.3.3. Bacteriophages. Another area of research is investigating attaching bacteriophages to implant surfaces. For this strategy, phage susceptibility testing must be done as phages have a narrow spectrum of activity [45]. Strengths of bacteriophage-implants include the limited ability of bacteria to develop resistance, autodosing depending on number of bacterial targets, low toxicity, minimal disruption of the commensal flora, lack of cross-resistance to antibiotics, and biofilm clearance [125]. Bacteriophage-coated implants are at an early clinical stage.

4.3. Strategies to disrupt biofilm architecture

4.3.1. Biofilm degrading enzymes

Certain enzymes are capable of cleaving and disrupting biofilm EPS matrix. These enzymes can be attached to coating surfaces to inhibit biofilm formation. For example, Dispersin B is bacterial enzyme able to degrade poly-N-acetylglucosamine (pNAG), which are a component of the biofilm matrix [126]. A coating has been developed via a layer-by-layer deposition of Dispersin B on the surface of a polymer [126]. The in vitro study demonstrated that Dispersin B coating was able to inhibit *S. epidermidis* biofilm formation [126]. Other glycosidases being investigated for their enzymatic activity against biofilms include alginate lyase, amylases, cellulases, and N-glycanases [127]. Another enzyme that is being examined for its biofilm disrupting capability is DNAse I which is able to degrade extracellular biofilm DNA [128]. Coating with DNase I has been shown to reduce bacterial adhesion and biofilm formation, without affecting mammalian cell adhesion and proliferation. [128]. A unique property of DNAse I is its ability to degrade biofilms from a wide range of bacterial species. Proteases are also able to disrupt biofilms, however these are less studied. An example of a protease capable of biofilm disruption is subtilisins, a serine protease commonly used in industry [127]. These strategies are currently in early pre-clinical stages.

4.3.2. Quorum sensing quenching

Bacterial cells communicate both intra-species and inter-species via quorum signalling molecules. Using quorum sensing, bacteria can orchestrate the development and expansion of biofilm EPS matrix [129]. Quorum sensing quenching enzymes can inhibit bacterial communication and disrupt biofilm formation. One study

developed a coating using PEG-based coating with covalent incorporation of a quorum sensing inhibitor, 5-Methylene-1-(prop-2-enoyl)-4-(2-fluorophen-yl)-dihydropyrrol-2-one (DHP), resulting in reduced cell attachment and biofilm formation [130]. Quorum sensing quenching strategies are currently in development for various medical devices such as catheters, dressings, contact lenses, and implantable devices [131]. These strategies are currently in early pre-clinical stages.

5. Comparison of anti-microbial coating strategies

Although significant advances and innovations have been made in the field of prosthesis coatings, currently the literature is lacking in quantitative comparative studies assessing the efficacy, toxicity, and durability of major antimicrobial coatings against each other. Furthermore, there is a paucity of clinical evidence for many of the previously discussed coatings. In this section, we will attempt to briefly compare some anti-microbial classes with the limited amount of comparative literature available and provide some recommendations for future study design.

Generally, in-organic coatings are more stable and less likely to induce anti-microbial resistance as compared to organic compounds. However, this usually comes at the expense of increased local toxicity to human tissue and potentially reduced osseointegration. As discussed later, the solution may be a synergistic and combinatorial application of these two coating classes.

Active and passive antimicrobial coatings strategies have been compared in the context of central venous catheters (CVC), in vitro [132]. While active antimicrobial coating strategies have shown more broad-spectrum anti-microbial activity, in the context of clinically relevant organisms comparable activity against gram-positive, gram-negative bacteria, and Candida species, have been observed by both coatings [132].

Another critical concern is the durability of coatings. Non-covalently adsorbed antibacterial coatings are chemically more stable but less firm and are more likely to separate from the coatings. Whereas chemical coating methods that covalently bond antibiotics to implant surfaces lose efficacy over time and become less stable [133]. As such, attempts are being made to create implant coating surfaces that have long-lasting renewable antibacterial efficacy with robust stability and biocompatibility [133]

Going forward, comparative studies need to assess coating antibacterial efficacy, osseointegration potential, and durability for various coatings classes. Therefore, standardized assays to compare coatings are required. For example, some well-studied *in vitro* assays that can be utilized to compare antibacterial efficacy of various coatings include: [134]

1. MTT (3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyl-2H-tetrazolium bromide) Assay
2. Optical Density (OD) 600 Assay
3. Disk diffusion assay
4. Colony Formation with Incubation in Solution Assay
5. Colony Formation on Soaked Coated Disks

For a review on standardised antibacterial material testing methods for coatings see Cunliffe et al. (2021) [135].

6. Current clinical outcomes

Despite adaptions to prosthesis design, surgical technique and operating room design, the rate of infection has remained relatively static. Although there are many potential areas for prosthesis surface treatment, as highlighted above, much of the literature evaluating clinical outcomes of anti-bacterial and anti-biofilm megaprosthetic coatings is focused on silver. Silver-coated megaprostheses were first used by Hardes (2006) in an attempt to reduce infection rates following LSS. These reports suggested that silver-coated megaprostheses reduced PJI risk, without causing toxic serum levels of silver (56.4 parts per billion) [87]. In addition, it was shown that there were no signs of local foreign body granulomatous reaction and no signs of systemic toxicity, with normal hepatic and renal function tests [87]. The same group also performed a 5 year prospective study and demonstrated that silver-coated prostheses reduced the infection rate in the medium term compared to a historical cohort [136]. Moreover, when infections occurred, management was made easier with silver-coated prostheses compared to the control group [136]. This is a key finding that requires validation, as treatment of PJI with irrigation and implant retention is significantly less burdensome to the patient and healthcare system than complete prosthesis removal and staged exchange. Other studies have identified that the activity of the silver-coating persisted for up to three years, as demonstrated by serum silver levels [137]. However, the tolerance of silver prostheses has been in question as there are some reports regarding potential consequences, including dermal argyria, ocular argyrosis, gastroenteritis and/or fever [138,139].

Of note, a large study of 394 patients found no significant difference between infection rates of silver-coated prostheses and uncoated prostheses, when utilized in high-risk patients in standard sites in primary bone tumours of the extremities. This may mean that surface treatment with silver “normalizes” the risk of infection in high-risk patients to those of low to normal risk, or it may reflect that there is no benefit, necessitating the need for more effective anti-microbial strategies. As the overall number of patients receiving megaprostheses whom develop infection is low, large cohorts of patients will be needed to achieve adequate statistical power. Table 3 displays some of the major clinical studies comparing PJIs in silver coated megaprostheses to uncoated controls.

Another surface treatment that has been tried clinically on megaprostheses is with an antibiotic-loaded hydrogel coating (Defensive Antibacterial Coating – DAC®) in 39 oncological patients and 3 non-oncologic patients. The hydrogel provided a reduction in early surgical site infections without any side effects [150]. DAC® hydrogel has also been used in combination with a bacteriophage for the treatment of a case of catastrophic relapsing *S. aureus* knee megaprosthesis infection [45]. Although ultimately the patient required an amputation as a result of other complications, the local infection control achieved by the DAC® hydrogel appeared favorable [45]. This case study also demonstrated the feasibility of phage-based coatings [45].

7. An evolutionary lens

Since the first eukaryotic cells evolved two billion years ago, we have been in an evolutionary arms race with bacteria [151]. There is a rapid emergence of multi-drug resistant bacteria around the world, which endangers antibiotic efficacy [152]. Additionally, there has been a higher prevalence of antibiotic-resistant organisms in the setting of PJIs [153]. Therefore innovative strategies to treat and prevent PJIs are necessary. However, an important consideration is that the use of a single strategy to fight bacterial infections applies an insufficient selective pressure on bacteria and allows for the evolution of resistant mutants. For example, bacteria have evolved various strategies to resist toxic metals, including reduced uptake, efflux pumps, extracellular and intracellular sequestration strategies, cellular repair, metabolic bypass, and chemical modification of metals [76].

Utilizing multimodal prevention and therapeutic strategies that do not function through the same mechanism of action have the highest potential to terminate bacteria, without allowing for resistant strains to evolve. We recommend future coatings utilize some combination of anti-adhesion strategies, active bactericidal strate-

Table 3
Studies Evaluating Clinical Outcomes on silver coated megaprostheses.

Study	# Patients	Total Rates of PJIs with Silver Coating	Total Rates of PJI in Uncoated Control	p -value	Mean Follow Up
Hardes [136]	51	5.9%	17.6%	0.062	19 months
Hussmann [140]	18	5.6%	22%	0.01	12 months
Wafa [141]	50	11.8%	22.4%	0.033	>12 months
Piccioli [142]	17	11.8%	23.1%	-	40.7 months
Donati [143]	38	7.9%	16.7%	-	46.5 months
Hardes [144]	56	8.9%	16.7%	0.247	38 months
Zajonz [145]	34	40%	57%	0.34	72.8 months
Streitbuerger [146]	64	9.4%	14.3%	-	34.5 months
Medellin [147]	81	17.4%	19%	0. 869	10.3 years
Parry [148]	394	12.4%	7.5%	0.154	55 months
Sambri [149]	29	10.3%	17.5%	0.104	36 months

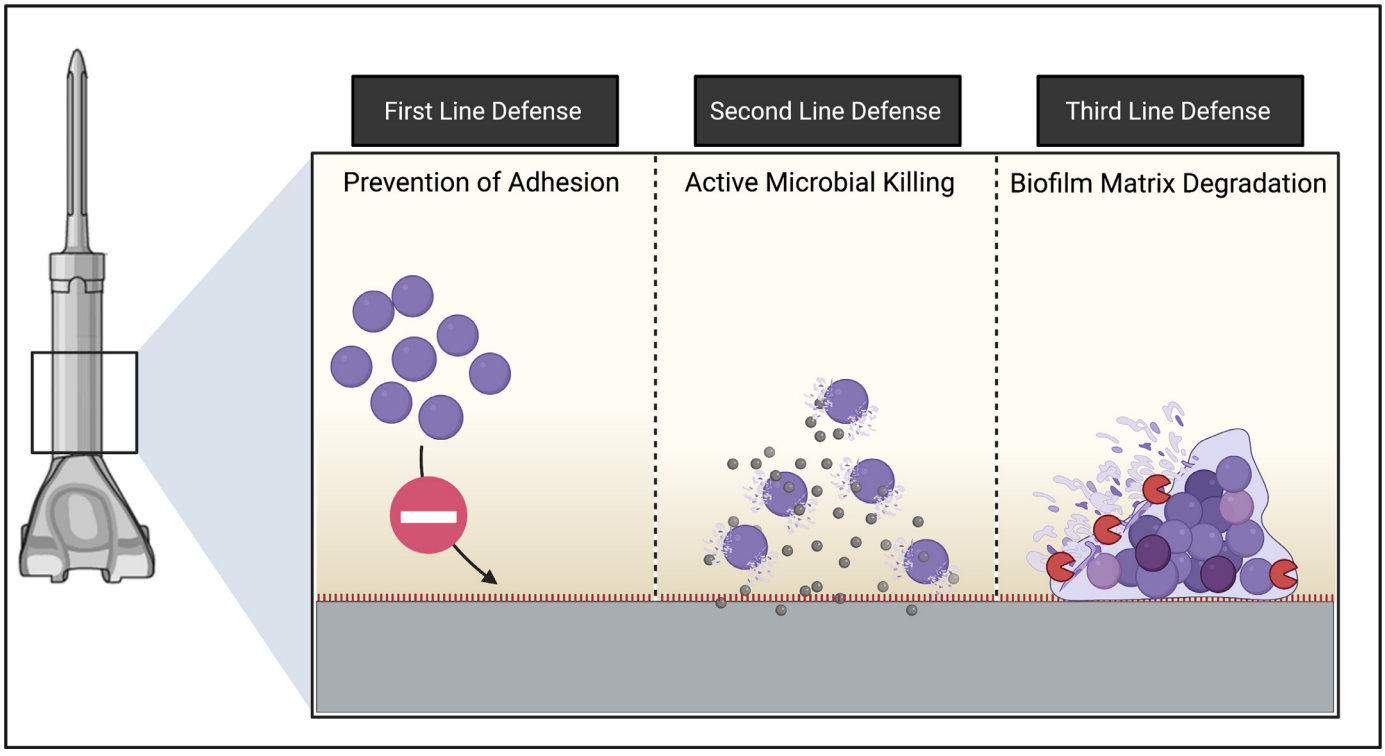

Fig. 6. Megaprosthesis with combination anti-microbial coating strategies.

gies, and biofilm disruption methods to prevent PJIs (Fig. 6). This can be thought of as a multistep defense line against biofilm formation [154]. While this type of coating system is scarce, efforts have been made in this regard. Some groups have attempted to create polymeric coating backbones that are non-adhesive, preventing conditioning film formation and bacterial adhesion (first line defense), while being able to be further functionalized with small peptide motifs (RGD) that promote osteoblast adhesion or modified to integrate bactericidal releasing agents (second line defense) or biofilm disrupting agents (third line defense) on top of it surface [154–157]. Therefore the solution to a combinatorial approach may be non-adhesive surfaces that are capable of being functionalized with further chemical groups and moieties, to achieve second and third line defenses.

8. Clinical and economic importance and future directions

Preventing infection following implantation of an orthopaedic implant is a large and important area of ongoing research. Clinicians and scientists have been focusing on this area due to the significant effect infections have on patients and the healthcare system. Estimates from the United States calculated that PJIs accounted for $1.62 billion in hospital costs [158]. This is largely due to the requirement for at least one subsequent operation, increasing costs by a factor of five [159]. Therefore, advances in implant technology may be significantly cost-effective if they are able to be implemented affordably. Due to the concerns for evolving bacterial resistance with liberal use of anti-microbials, a combination approach should be considered. However, use of combination strategies must also consider toxicity to the patient as well as associated costs. As described earlier there is a major trade-off between antibacterial coating's bactericidal efficacy and local eukaryotic cytotoxicity, therefore it is critical to utilize multimodal strategies that while disrupting bacterial life cycle and adhesion, have minimal effects on the human host cells. Furthermore, the anti-bacterial coating should not impair osteoblast function and effective osseointegration.

Regarding all antimicrobial prosthesis coatings, at present the most used strategy is bone cement loaded with antibiotics. However, the quality of available clinical evidence is poor and consensus from the most recent (2018) International Consensus on Orthopaedic Infections suggest it should only be considered in patients at high risk of infection [122]. Patients undergoing megaprosthesis implantation following a bone tumour resection

are considered at high risk of infection. There is an ongoing randomized controlled trial (RCT) currently evaluating the effectiveness of antibiotic loaded cement (NCT04135170). We were also able to identify an ongoing trial evaluating the effectiveness of single-stage revision surgery with DAC® hydrogel compared to traditional two-stage revision without this antimicrobial coating for the prevention of re-infection following PJI (NCT04251377). Custom iodine-coated prostheses developed by a team in Japan have also shown promising results, with a 4.2% rate of re-infection following their implantation. Various implant types were coated and used in different anatomical regions to prevent secondary infections [160,161].

Several clinical questions remain, for which there is a current paucity of literature. Determining the duration that antimicrobial coatings remain at a therapeutic level on the implant is important. While some coatings may be effective at preventing infection and biofilm formation *in vitro*, antimicrobial duration *in vivo* needs to persist long enough to prevent the development of infection. Some coatings may perform long enough to prevent acute infections, but not chronic infections. Moreover, some coatings act broadly, passively preventing against infection from a wide range of bacterial pathogens, whereas some coatings are more specific, only activated in the presence of a biofilm, rendering them most effective against certain biofilm-producing pathogens (for example, *Enterococcus, Staphylococcus spp., Streptococcus and Pseudomonas).* Therefore, in the setting of revision surgery for PJI, the specific pathogen may be a consideration when selecting a coated prosthesis. The clinical indications and risk factors to consider for inserting a coated prosthesis need to be determined for all patients receiving arthroplasty-type prostheses. Patients undergoing megaprothesis reconstruction are at significantly higher risk of infection than the typical joint replacement cohort, therefore, we believe that ongoing efforts to ascertain the optimal coating for reducing PJIs in megaprostheses are essential.

Overall, this review highlighted the multitude of potential solutions to combating PJIs in megaprotheses. Results of clinical studies evaluating coated prostheses are encouraging, but further research and technological developments are required. We look forward to the results of the ongoing clinical trials for each of the antimicrobial strategies mentioned. As different coatings become available on the market, direct comparison between implant-types and determining effectiveness over time and against specific bacteria will be essential. Reducing infection rates following the implantation of megaprostheses would have a significant impact on sarcoma care and major trauma surgeries that require reconstruction of large skeletal defects.

Financial support

There was no funding or financial support utilized to carry out this research.

Disclosures

Each author certifies that he or she has no commercial associations (e.g. Consultancies, stock ownership, equity interest, patent/licensing arrangements, etc.) that might pose a conflict of interest in connection with the submitted article.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Acknowledgement

In this review Figs. 1,2,4,5, and 6 were created by Biorender.com.

References

[1] H. Ozger, M. Bulbul, L. Eralp, Complications of limb salvage surgery in childhood tumors and recommended solutions, Strat. Trauma Limb Reconstr. 5 (2010) 11–15, doi:10.1007/s11751-009-0075-y.
[2] M.A. Ayerza, G.L. Farfalli, L. Aponte-Tinao, D.L. Muscolo, Does increased rate of limb-sparing surgery affect survival in osteosarcoma? Clin. Orthop. Relat. Res. 468 (2010) 2854–2859, doi:10.1007/s11999-010-1423-4.
[3] J. Stevenson, P. Tsagkozis, R. Grimer, Functional and quality of life outcomes in bone sarcoma following amputation, rotationplasty or limb-salvage, Expert Rev. Qual. Life Cancer Care 1 (2016) 303–312, doi:10.1080/23809000.2016.1203725.
[4] S. Furtado, R.J. Grimer, P. Cool, S.A. Murray, T. Briggs, J. Fulton, K. Grant, C.H. Gerrand, Physical functioning, pain and quality of life after amputation for musculoskeletal tumours: a national survey, Bone Joint J. 97-B (2015) 1284–1290, doi:10.1302/0301-620X.97B9.35192.
[5] F. Malek, J.S. Somerson, S. Mitchel, R.P. Williams, Does limb-salvage surgery offer patients better quality of life and functional capacity than amputation? Clin. Orthop. Relat. Res. 470 (2012) 2000–2006, doi:10.1007/s11999-012-2271-1.
[6] M.A. Simon, M.A. Aschliman, N. Thomas, H.J. Mankin, Limb-salvage treatment versus amputation for osteosarcoma of the distal end of the femur. 1986, J. Bone Joint Surg. Am. 87 (2005) 2822, doi:10.2106/JBJS.8712.cl.
[7] G. Han, W.-Z. Bi, M. Xu, J.-P. Jia, Y. Wang, Amputation versus limb-salvage surgery in patients with osteosarcoma: a meta-analysis, World J. Surg. 40 (2016) 2016–2027, doi:10.1007/s00268-016-3500-7.
[8] L.M. Jeys, R.J. Grimer, S.R. Carter, R.M. Tillman, Periprosthetic infection in patients treated for an orthopaedic oncological condition, J. Bone Joint Surg. Am. 87 (2005) 842–849, doi:10.2106/JBJS.C.01222.
[9] H.-M. Peng, L.-C. Wang, J.-Y. Cheng, Y.-X. Zhou, H. Tian, J.-H. Lin, W.-S. Guo, Y. Lin, T.-B. Qu, A. Guo, Y.-P. Cao, X.-S. Weng, Rates of periprosthetic infection and surgical revision in Beijing (China) between 2014 and 2016: a retrospective multicenter cross-sectional study, J. Orthop. Surg. Res. 14 (2019) 463, doi:10.1186/s13018-019-1520-3.
[10] R.-J.W.J. Zuidhof, C.A.M. Löwik, J.J.W. Ploegmakers, S.P.D. Dijkstra, M. Wouthuyzen-Bakker, P.C. Jutte, Periprosthetic joint infection in orthopaedic surgical oncology, Ann. Jt. 4 (2019) 26 –26, doi:10.21037/aoj.2019.05.01.
[11] T. Lin, Q. Jin, X. Mo, Z. Zhao, X. Xie, C. Zou, G. Huang, J. Yin, J. Shen, Experience with periprosthetic infection after limb salvage surgery for patients with osteosarcoma, J. Orthop. Surg. Res. 16 (2021) 93, doi:10.1186/s13018-021-02243-6.
[12] S. Nagano, M. Yokouchi, T. Setoguchi, H. Sasaki, H. Shimada, I. Kawamura, Y. Ishidou, J. Kamizono, T. Yamamoto, H. Kawamura, S. Komiya, Analysis of surgical site infection after musculoskeletal tumor surgery: risk assessment using a new scoring system, Sarcoma 2014 (2014) 645496, doi:10.1155/2014/645496.
[13] A. Racano, T. Pazionis, F. Farrokhyar, B. Deheshi, M. Ghert, High infection rate outcomes in long-bone tumor surgery with endoprosthetic reconstruction in adults: a systematic review, Clin. Orthop. Relat. Res. 471 (2013) 2017–2027, doi:10.1007/s11999-013-2842-9.
[14] T. Peel, D. May, K. Buising, K. Thursky, M. Slavin, P. Choong, Infective complications following tumour endoprosthesis surgery for bone and soft tissue tumours, Eur. J. Surg. Oncol. 40 (2014) 1087–1094, doi:10.1016/j.ejso.2014.02.241.
[15] H. Pilge, G. Gradl, R. von Eisenhart-Rothe, H. Gollwitzer, Incidence and outcome after infection of megaprostheses, Hip Int. J. Clin. Exp. Res. Hip Pathol. Ther. 22 (Suppl 8) (2012) S83–S90, doi:10.5301/HIP.2012.9576.
[16] A.J. Tande, R. Patel, Prosthetic joint infection, Clin. Microbiol. Rev. 27 (2014) 302–345, doi:10.1128/CMR.00111-13.
[17] A. Shahi, J. Parvizi, Prevention of periprosthetic joint infection, Arch. Bone Jt. Surg. 3 (2015) 72–81.
[18] M. Ghert, B. Deheshi, G. Holt, R.L. Randall, P. Ferguson, J. Wunder, R. Turcotte, J. Werier, P. Clarkson, T. Damron, J. Benevenia, M. Anderson, M. Gebhardt, M. Isler, S. Mottard, J. Healey, N. Evaniew, A. Racano, S. Sprague, M. Swinton, D. Bryant, L. Thabane, G. Guyatt, M. Bhandari, Prophylactic antibiotic regimens in tumour surgery (PARITY): protocol for a multicentre randomised controlled study, BMJ Open 2 (2012), doi:10.1136/bmjopen-2012-002197.
[19] C.L. Romanò, H. Tsuchiya, I. Morelli, A.G. Battaglia, L. Drago, Antibacterial coating of implants: are we missing something? Bone Joint Res. 8 (2019) 199–206, doi:10.1302/2046-3758.85.BJR-2018-0316.
[20] D. Vidlak, T. Kielian, Infectious dose dictates the host response during Staphylococcus aureus orthopedic-implant biofilm infection, Infect. Immun. 84 (2016) 1957–1965, doi:10.1128/IAI.00117-16.
[21] S. Veerachamy, T. Yarlagadda, G. Manivasagam, P.K. Yarlagadda, Bacterial adherence and biofilm formation on medical implants: a review, Proc. Inst. Mech. Eng. Part H, J. Eng. Med. 228 (2014) 1083–1099, doi:10.1177/0954411914556137.

[22] Z. Khatoon, C.D. McTiernan, E.J. Suuronen, T.-F. Mah, E.I. Alarcon, Bacterial biofilm formation on implantable devices and approaches to its treatment and prevention, Heliyon 4 (2018) e01067, doi:10.1016/j.heliyon.2018.e01067.
[23] H. Kanematsu, D.M. Barry, Conditioning Films BT - Biofilm and Materials Science, in: H. Kanematsu, D.M. Barry (Eds.), Springer International Publishing, Cham, 2015: pp. 9–15. doi:10.1007/978-3-319-14565-5_2.
[24] J. Gallo, M. Holinka, C.S. Moucha, Antibacterial surface treatment for orthopaedic implants, Int. J. Mol. Sci. 15 (2014) 13849–13880, doi:10.3390/ijms150813849.
[25] P. Gupta, S. Sarkar, B. Das, S. Bhattacharjee, P. Tribedi, Biofilm, pathogenesis and prevention–a journey to break the wall: a review, Arch. Microbiol. 198 (2016) 1–15, doi:10.1007/s00203-015-1148-6.
[26] H.O. Gbejuade, A.M. Lovering, J.C. Webb, The role of microbial biofilms in prosthetic joint infections, Acta Orthop. 86 (2015) 147–158, doi:10.3109/17453674.2014.966290.
[27] J. Quayle, A. Barakat, A. Klasan, A. Mittal, G. Chan, J. Gibbs, M. Edmondson, P. Stott, Management of peri-prosthetic joint infection and severe bone loss after total hip arthroplasty using a long-stemmed cemented custom-made articulating spacer (CUMARS), BMC Musculoskelet. Disord. 22 (2021) 358, doi:10.1186/s12891-021-04237-1.
[28] T.K. Wood, Precedence for the structural role of Flagella in biofilms, MBio 4 (2013), doi:10.1128/mBio.00225-13.
[29] X. Jin, J.S. Marshall, Mechanics of biofilms formed of bacteria with fimbriae appendages, PLoS One 15 (2020) e0243280, doi:10.1371/journal.pone.0243280.
[30] C. Berne, A. Ducret, G.G. Hardy, Y.V Brun, Adhesins involved in attachment to abiotic surfaces by gram-negative bacteria, Microbiol. Spectr. 3 (2015), doi:10.1128/microbiolspec.MB-0018-2015.
[31] R.E. Kania, G.E.M. Lamers, M.J. Vonk, P.T.B. Huy, P.S. Hiemstra, G.V Bloemberg, J.J. Grote, Demonstration of bacterial cells and glycocalyx in biofilms on human tonsils, Arch. Otolaryngol. Head. Neck Surg. 133 (2007) 115–121, doi:10.1001/archotol.133.2.115.
[32] P. Di Martino, Extracellular polymeric substances, a key element in understanding biofilm phenotype, AIMS Microbiol. 4 (2018) 274–288, doi:10.3934/microbiol.2018.2.274.
[33] R.M. Donlan, Biofilms: microbial life on surfaces, Emerg. Infect. Dis. 8 (2002) 881–890, doi:10.3201/eid0809.020063.
[34] P. Saxena, Y. Joshi, K. Rawat, R. Bisht, Biofilms: architecture, resistance, quorum sensing and control mechanisms, Indian J. Microbiol. 59 (2019) 3–12, doi:10.1007/s12088-018-0757-6.
[35] K. Bisht, C.A. Wakeman, Discovery and therapeutic targeting of differentiated biofilm subpopulations, Front. Microbiol. 10 (2019) 1908, doi:10.3389/fmicb.2019.01908.
[36] S. Singh, S.K. Singh, I. Chowdhury, R. Singh, Understanding the mechanism of bacterial biofilms resistance to antimicrobial agents, Open Microbiol. J. 11 (2017) 53–62, doi:10.2174/1874285801711010053.
[37] T.F. Mah, G.A. O'Toole, Mechanisms of biofilm resistance to antimicrobial agents, Trends Microbiol. 9 (2001) 34–39, doi:10.1016/s0966-842x(00)01913-2.
[38] A.S. Wasterlain, K. Goswami, S.A. Ghasemi, J. Parvizi, Diagnosis of periprosthetic infection: recent developments, JBJS 102 (2020) https://journals.lww.com/jbjsjournal/subjects/Hip/Fulltext/2020/08050/Diagnosis_of_Periprosthetic_Infection__Recent.16.aspx.
[39] J. Parvizi, T.L. Tan, K. Goswami, C. Higuera, C.Della Valle, A.F. Chen, N. Shohat, The 2018 definition of periprosthetic hip and knee infection: an evidence-based and validated criteria, J. Arthroplasty 33 (2018) 1309–1314 e2, doi:10.1016/j.arth.2018.02.078.
[40] J.R. Lex, J. Gregory, C. Allen, J.P. Reid, J.D. Stevenson, Distinguishing bone and soft tissue infections mimicking sarcomas requires multimodal multidisciplinary team assessment, Ann. R. Coll. Surg. Engl. 101 (6) (2019) 405–410, doi:10.1308/rcsann.2019.0040.
[41] B.H. Kapadia, R. Pivec, A.J. Johnson, K. Issa, Q. Naziri, J.A. Daley, M.A. Mont, Infection prevention methodologies for lower extremity total joint arthroplasty, Expert Rev. Med. Devices 10 (2013) 215–224, doi:10.1586/erd.12.76.
[42] W. Zimmerli, A. Trampuz, P.E. Ochsner, Prosthetic-joint infections, N. Engl. J. Med. 351 (2004) 1645–1654, doi:10.1056/NEJMra040181.
[43] G. Vrgoc, M. Japjec, G. Gulan, J. Ravlić-Gulan, M. Marinović, A. Bandalović, Periprosthetic infections after total hip and knee arthroplasty–a review, Coll. Antropol. 38 (2014) 1259–1264.
[44] N.H. Søe, N.V. Jensen, A.L. Jensen, J. Koch, S.S. Poulsen, G.B. Pier, H.K. Johansen, Active and passive immunization against Staphylococcus aureus periprosthetic osteomyelitis in rats, In Vivo 31 (2017) 45–50, doi:10.21873/invivo.11023.
[45] T. Ferry, C. Batailler, C. Petitjean, J. Chateau, C. Fevre, E. Forestier, S. Brosset, G. Leboucher, C. Kolenda, F. Laurent, S. Lustig, The potential innovative use of bacteriophages within the DAC(®) hydrogel to treat patients with knee megaprosthesis infection requiring "debridement antibiotics and implant retention" and soft tissue coverage as salvage therapy, Front. Med. 7 (2020) 342, doi:10.3389/fmed.2020.00342.
[46] O.D. Savvidou, A. Kaspiris, I. Trikoupis, G. Kakouratos, S. Goumenos, D. Melissaridou, P.J. Papagelopoulos, Efficacy of antimicrobial coated orthopaedic implants on the prevention of periprosthetic infections: a systematic review and meta-analysis, J. Bone Jt. Infect. 5 (2020) 212–222, doi:10.7150/jbji.44839.
[47] K.G. Neoh, E.T. Kang, Combating bacterial colonization on metals via polymer coatings: relevance to marine and medical applications, ACS Appl. Mater. Interfaces 3 (2011) 2808–2819, doi:10.1021/am200646t.
[48] I. Francolini, L. Hall-Stoodley, P. Stoodley, 2.2.8 - Biofilms, biomaterials, and device-related infections, in: W.R. Wagner, S.E. Sakiyama-Elbert, G. Zhang, M.J. Yaszemski (Eds.), Biomater. Sci. (Fourth Ed., Fourth Edi, Academic Press, 2020, pp. 823–840, doi:10.1016/B978-0-12-816137-1.00054-4.
[49] S.B. Goodman, Z. Yao, M. Keeney, F. Yang, The future of biologic coatings for orthopaedic implants, Biomaterials 34 (2013) 3174–3183, doi:10.1016/j.biomaterials.2013.01.074.
[50] G. Gosheger, C. Goetze, J. Hardes, U. Joosten, W. Winkelmann, C. von Eiff, The influence of the alloy of megaprostheses on infection rate, J. Arthroplasty 23 (2008) 916–920, doi:10.1016/j.arth.2007.06.015.
[51] D.W. Howie, S.D. Rogers, M.A. McGee, D.R. Haynes, Biologic effects of cobalt chrome in cell and animal models, Clin. Orthop. Relat. Res. (1996) S217–S232, doi:10.1097/00003086-199608001-00019.
[52] A. Shanbhag, J. Yang, J. Lilien, J. Black, Decreased neutrophil respiratory burst on exposure to cobalt-chrome alloy and polystyrene in vitro, J. Biomed. Mater. Res. 26 (1992) 185–195, doi:10.1002/jbm.820260205.
[53] S.S. Patel, W. Aruni, S. Inceoglu, Y.T. Akpolat, G.D. Botimer, W.K. Cheng, O.A. Danisa, A comparison of Staphylococcus aureus biofilm formation on cobalt-chrome and titanium-alloy spinal implants, J. Clin. Neurosci. Off. J. Neurosurg. Soc. Australas. 31 (2016) 219–223, doi:10.1016/j.jocn.2016.03.013.
[54] K. Watanabe, S. Fukuzaki, A. Sugino, N. Benson, N. Metcalf, M. Nakamura, M. Matsumoto, Cobalt–chromium alloy has superior antibacterial effect than titanium alloy: in vitro and in vivo studies, Spine (Phila. Pa. 1976) 46 (2021) https://journals.lww.com/spinejournal/Fulltext/2021/09010/Cobalt_Chromium_Alloy_Has_Superior_Antibacterial.4.aspx.
[55] X. Khoo, G.A. O'Toole, S.A. Nair, B.D. Snyder, D.J. Kenan, M.W. Grinstaff, Staphylococcus aureus resistance on titanium coated with multivalent PEGylated-peptides, Biomaterials 31 (2010) 9285–9292, doi:10.1016/j.biomaterials.2010.08.031.
[56] X. Khoo, P. Hamilton, G.A. O'Toole, B.D. Snyder, D.J. Kenan, M.W. Grinstaff, Directed assembly of PEGylated-peptide coatings for infection-resistant titanium metal, J. Am. Chem. Soc. 131 (2009) 10992–10997, doi:10.1021/ja9020827.
[57] L.G. Harris, L.M. Patterson, C. Bacon, I. ap Gwynn, R.G. Richards, Assessment of the cytocompatibility of different coated titanium surfaces to fibroblasts and osteoblasts, J. Biomed. Mater. Res. A 73 (2005) 12–20, doi:10.1002/jbm.a.30276.
[58] G. Subbiahdoss, B. Pidhatika, G. Coullerez, M. Charnley, R. Kuijer, H.C. van der Mei, M. Textor, H.J. Busscher, Bacterial biofilm formation versus mammalian cell growth on titanium-based mono- and bi-functional coating, Eur. Cell. Mater. 19 (2010) 205–213, doi:10.22203/ecm.v019a20.
[59] P.-H. Chua, K.-G. Neoh, E.-T. Kang, W. Wang, Surface functionalization of titanium with hyaluronic acid/chitosan polyelectrolyte multilayers and RGD for promoting osteoblast functions and inhibiting bacterial adhesion, Biomaterials 29 (2008) 1412–1421, doi:10.1016/j.biomaterials.2007.12.019.
[60] M. Kong, X.G. Chen, K. Xing, H.J. Park, Antimicrobial properties of chitosan and mode of action: a state of the art review, Int. J. Food Microbiol. 144 (2010) 51–63, doi:10.1016/j.ijfoodmicro.2010.09.012.
[61] M. Hosseinnejad, S.M. Jafari, Evaluation of different factors affecting antimicrobial properties of chitosan, Int. J. Biol. Macromol. 85 (2016) 467–475, doi:10.1016/j.ijbiomac.2016.01.022.
[62] R.R. Maddikeri, S. Tosatti, M. Schuler, S. Chessari, M. Textor, R.G. Richards, L.G. Harris, Reduced medical infection related bacterial strains adhesion on bioactive RGD modified titanium surfaces: a first step toward cell selective surfaces, J. Biomed. Mater. Res. A. 84 (2008) 425–435, doi:10.1002/jbm.a.31323.
[63] M. Chai, M. An, X. Zhang, Construction of a TiO(2)/MoSe(2)/CHI coating on dental implants for combating Streptococcus mutans infection, Mater. Sci. Eng. C. Mater. Biol. Appl. 129 (2021) 112416, doi:10.1016/j.msec.2021.112416.
[64] M. Franceschini, N.A. Sandiford, V. Cerbone, L.C.T. de Araujo, D. Kendoff, Defensive antibacterial coating in revision total hip arthroplasty: new concept and early experience, Hip Int. J. Clin. Exp. Res. Hip Pathol. Ther. 30 (2020) 7–11, doi:10.1177/1120700020917125.
[65] K. Malizos, M. Blauth, A. Danita, N. Capuano, R. Mezzoprete, N. Logoluso, L. Drago, C.L. Romanò, Fast-resorbable antibiotic-loaded hydrogel coating to reduce post-surgical infection after internal osteosynthesis: a multicenter randomized controlled trial, J. Orthop. Traumatol. Off. J. Ital. Soc. Orthop. Traumatol. 18 (2017) 159–169, doi:10.1007/s10195-017-0442-2.
[66] A.M. Gallardo-Moreno, M.A. Pacha-Olivenza, L. Saldaña, C. Pérez-Giraldo, J.M. Bruque, N. Vilaboa, M.L. González-Martín, In vitro biocompatibility and bacterial adhesion of physico-chemically modified Ti6Al4V surface by means of UV irradiation, Acta Biomater. 5 (2009) 181–192, doi:10.1016/j.actbio.2008.07.028.
[67] J.C. Yu, W. Ho, J. Lin, H. Yip, P.K. Wong, Photocatalytic activity, antibacterial effect, and photoinduced hydrophilicity of TiO2 films coated on a stainless steel substrate, Environ. Sci. Technol. 37 (2003) 2296–2301, doi:10.1021/es0259483.
[68] A. Jaggessar, H. Shahali, A. Mathew, P.K.D.V Yarlagadda, Bio-mimicking nano and micro-structured surface fabrication for antibacterial properties in medical implants, J. Nanobiotechnol. 15 (2017) 64, doi:10.1186/s12951-017-0306-1.
[69] S. Wu, B. Zhang, Y. Liu, X. Suo, H. Li, Influence of surface topography on bacterial adhesion: a review (Review), Biointerphases 13 (2018) 60801, doi:10.1116/1.5054057.
[70] P. Colson, C. Henrist, R. Cloots, Nanosphere lithography: a powerful method for the controlled manufacturing of nanomaterials, J. Nanomater. 2013 (2013) 948510, doi:10.1155/2013/948510.

[71] P. Tang, W. Zhang, Y. Wang, B. Zhang, H. Wang, C. Lin, L. Zhang, Effect of superhydrophobic surface of titanium on *Staphylococcus aureus* adhesion, J. Nanomater. 2011 (2011) 178921, doi:10.1155/2011/178921.
[72] B. Del Curto, M.F. Brunella, C. Giordano, M.P. Pedeferri, V. Valtulina, L. Visai, A. Cigada, Decreased bacterial adhesion to surface-treated titanium, Int. J. Artif. Organs. 28 (2005) 718–730, doi:10.1177/039139880502800711.
[73] L. Rodrigues, I.M. Banat, J. Teixeira, R. Oliveira, Biosurfactants: potential applications in medicine, J. Antimicrob. Chemother. 57 (2006) 609–618, doi:10.1093/jac/dkl024.
[74] F. Rivardo, R.J. Turner, G. Allegrone, H. Ceri, M.G. Martinotti, Anti-adhesion activity of two biosurfactants produced by Bacillus spp. prevents biofilm formation of human bacterial pathogens, Appl. Microbiol. Biotechnol. 83 (2009) 541–553, doi:10.1007/s00253-009-1987-7.
[75] E. Tambone, E. Bonomi, P. Ghensi, D. Maniglio, C. Ceresa, F. Agostinacchio, P. Caciagli, G. Nollo, F. Piccoli, I. Caola, L. Fracchia, F. Tessarolo, Rhamnolipid coating reduces microbial biofilm formation on titanium implants: an in vitro study, BMC Oral Health 21 (2021) 49, doi:10.1186/s12903-021-01412-7.
[76] J.A. Lemire, J.J. Harrison, R.J. Turner, Antimicrobial activity of metals: mechanisms, molecular targets and applications, Nat. Rev. Microbiol. 11 (2013) 371–384, doi:10.1038/nrmicro3028.
[77] R.M. Lopachin, T. Gavin, A. Decaprio, D.S. Barber, Application of the Hard and Soft, Acids and Bases (HSAB) theory to toxicant–target interactions, Chem. Res. Toxicol. 25 (2012) 239–251, doi:10.1021/tx2003257.
[78] A. Diez-Escudero, N.P. Hailer, The role of silver coating for arthroplasty components, Bone Joint J 103-B (2021) 423–429, doi:10.1302/0301-620X.103B3.BJJ-2020-1370.R1.
[79] T.C. Dakal, A. Kumar, R.S. Majumdar, V. Yadav, Mechanistic basis of antimicrobial actions of silver nanoparticles, Front. Microbiol. 7 (2016) 1831, doi:10.3389/fmicb.2016.01831.
[80] I.X. Yin, J. Zhang, I.S. Zhao, M.L. Mei, Q. Li, C.H. Chu, The antibacterial mechanism of silver nanoparticles and its application in dentistry, Int. J. Nanomedicine. 15 (2020) 2555–2562, doi:10.2147/IJN.S246764.
[81] T. Schmidt-Braekling, A. Streitbuerger, G. Gosheger, F. Boettner, M. Nottrott, H. Ahrens, R. Dieckmann, W. Guder, D. Andreou, G. Hauschild, B. Moellenbeck, W. Waldstein, J. Hardes, Silver-coated megaprostheses: review of the literature, Eur. J. Orthop. Surg. Traumatol. 27 (2017) 483–489, doi:10.1007/s00590-017-1933-9.
[82] J.L. Clement, P.S. Jarrett, Antibacterial silver, Met. Based. Drugs. 1 (1994) 467–482, doi:10.1155/MBD.1994.467.
[83] O. Choi, K.K. Deng, N.-J. Kim, L. Ross, R.Y. Surampalli, Z. Hu, The inhibitory effects of silver nanoparticles, silver ions, and silver chloride colloids on microbial growth, Water Res. 42 (2008) 3066–3074, doi:10.1016/j.watres.2008.02.021.
[84] M.C. Wyatt, M. Foxall-Smith, A. Roberton, A. Beswick, D.C. Kieser, M.R. Whitehouse, The use of silver coating in hip megaprostheses: a systematic review, Hip Int. J. Clin. Exp. Res. Hip Pathol. Ther. 29 (2019) 7–20, doi:10.1177/1120700018811070.
[85] S. Eto, S. Kawano, S. Someya, H. Miyamoto, M. Sonohata, M. Mawatari, First clinical experience with thermal-sprayed silver oxide-containing hydroxyapatite coating implant, J. Arthroplasty 31 (2016) 1498–1503, doi:10.1016/j.arth.2015.12.034.
[86] G.V Morris, J. Kozdryk, J. Gregory, L. Jeys, The use of silver-coated orthopaedic implants: are all silvers the same? Curr. Orthop. Pract. 28 (2017) https://journals.lww.com/c-orthopaedicpractice/Fulltext/2017/11000/The_use_of_silver_coated_orthopaedic_implants__are.5.aspx.
[87] J. Hardes, H. Ahrens, C. Gebert, A. Streitbuerger, H. Buerger, M. Erren, A. Gunsel, C. Wedemeyer, G. Saxler, W. Winkelmann, G. Gosheger, Lack of toxicological side-effects in silver-coated megaprostheses in humans, Biomaterials 28 (2007) 2869–2875, doi:10.1016/j.biomaterials.2007.02.033.
[88] D.-H. Song, S.-H. Uhm, S.-B. Lee, J.-G. Han, K.-N. Kim, Antimicrobial silver-containing titanium oxide nanocomposite coatings by a reactive magnetron sputtering, Thin Solid Films 519 (2011) 7079–7085, doi:10.1016/j.tsf.2011.01.385.
[89] A. Ewald, S.K. Glückermann, R. Thull, U. Gbureck, Antimicrobial titanium/silver PVD coatings on titanium, Biomed. Eng. Online 5 (2006) 22, doi:10.1186/1475-925X-5-22.
[90] F. Boccafoschi, L. Fusaro, M. Cannas, 15 - Immobilization of Peptides on Cardiovascular Stent, in: J.G. Wall, H. Podbielska, M.B.T.-F.C.S. Wawrzyńska (Eds.), Woodhead Publishing, 2018: pp. 305–318. doi:10.1016/B978-0-08-100496-8.00016-0.
[91] L. Juan, Z. Zhimin, M. Anchun, L. Lei, Z. Jingchao, Deposition of silver nanoparticles on titanium surface for antibacterial effect, Int. J. Nanomed. 5 (2010) 261–267, doi:10.2147/ijn.s8810.
[92] X. Zeng, S. Xiong, S. Zhuo, C. Liu, J. Miao, D. Liu, H. Wang, Y. Zhang, C. Wang, Y. Liu, Nanosilver/poly (dl-lactic-co-glycolic acid) on titanium implant surfaces for the enhancement of antibacterial properties and osteoinductivity, Int. J. Nanomed. 14 (2019) 1849–1863, doi:10.2147/IJN.S190954.
[93] R. Kuehl, P.S. Brunetto, A.-K. Woischnig, M. Varisco, Z. Rajacic, J. Vosbeck, L. Terracciano, K.M. Fromm, N. Khanna, Preventing implant-associated infections by silver coating, Antimicrob. Agents Chemother. 60 (2016) 2467–2475, doi:10.1128/AAC.02934-15.
[94] M. Shimabukuro, Antibacterial property and biocompatibility of silver, copper, and zinc in titanium dioxide layers incorporated by one-step micro-arc oxidation: a review, Antibiot. (Basel, Switzerland) 9 (2020), doi:10.3390/antibiotics9100716.
[95] M.V Sofroniew, H.V Vinters, Astrocytes: biology and pathology, Acta Neuropathol. 119 (2010) 7–35, doi:10.1007/s00401-009-0619-8.
[96] P. Petrini, C.R. Arciola, I. Pezzali, S. Bozzini, L. Montanaro, M.C. Tanzi, P. Speziale, L. Visai, Antibacterial activity of zinc modified titanium oxide surface, Int. J. Artif. Organs. 29 (2006) 434–442, doi:10.1177/039139880602900414.
[97] G. Jin, H. Cao, Y. Qiao, F. Meng, H. Zhu, X. Liu, Osteogenic activity and antibacterial effect of zinc ion implanted titanium, Colloids Surf. B. Biointerfaces 117 (2014) 158–165, doi:10.1016/j.colsurfb.2014.02.025.
[98] H. Hu, W. Zhang, Y. Qiao, X. Jiang, X. Liu, C. Ding, Antibacterial activity and increased bone marrow stem cell functions of Zn-incorporated TiO2 coatings on titanium, Acta Biomater. 8 (2012) 904–915, doi:10.1016/j.actbio.2011.09.031.
[99] G. Grass, C. Rensing, M. Solioz, Metallic copper as an antimicrobial surface, Appl. Environ. Microbiol. 77 (2011) 1541–1547, doi:10.1128/AEM.02766-10.
[100] C. Bergemann, S. Zaatreh, K. Wegner, K. Arndt, A. Podbielski, R. Bader, C. Prinz, U. Lembke, J.B. Nebe, Copper as an alternative antimicrobial coating for implants—An in vitro study, World J. Transplant. 7 (2017) 193–202, doi:10.5500/wjt.v7.i3.193.
[101] V. Hodgkinson, M.J. Petris, Copper homeostasis at the host-pathogen interface, J. Biol. Chem. 287 (2012) 13549–13555, doi:10.1074/jbc.R111.316406.
[102] T. Shirai, H. Tsuchiya, T. Shimizu, K. Ohtani, Y. Zen, K. Tomita, Prevention of pin tract infection with titanium-copper alloys, J. Biomed. Mater. Res. B. Appl. Biomater. 91 (2009) 373–380, doi:10.1002/jbm.b.31412.
[103] F. Heidenau, W. Mittelmeier, R. Detsch, M. Haenle, F. Stenzel, G. Ziegler, H. Gollwitzer, A novel antibacterial titania coating: metal ion toxicity and in vitro surface colonization, J. Mater. Sci. Mater. Med. 16 (2005) 883–888, doi:10.1007/s10856-005-4422-3.
[104] J.A. Garza-Cervantes, A. Chávez-Reyes, E.C. Castillo, G. García-Rivas, O.Antonio Ortega-Rivera, E. Salinas, M. Ortiz-Martínez, S.L. Gómez-Flores, J.A. Peña-Martínez, A. Pepi-Molina, M.T. Treviño-González, X. Zarate, M.Elena Cantú-Cárdenas, C.Enrique Escarcega-Gonzalez, J.R. Morones-Ramírez, Synergistic antimicrobial effects of silver/transition-metal combinatorial treatments, Sci. Rep. 7 (2017) 903, doi:10.1038/s41598-017-01017-7.
[105] G. Cottarel, J. Wierzbowski, Combination drugs, an emerging option for antibacterial therapy, Trends Biotechnol. 25 (2007) 547–555, doi:10.1016/j.tibtech.2007.09.004.
[106] K. Ueoka, T. Kabata, M. Tokoro, Y. Kajino, D. Inoue, T. Takagi, T. Ohmori, J. Yoshitani, T. Ueno, Y. Yamamuro, A. Taninaka, H. Tsuchiya, Antibacterial activity in iodine-coated implants under conditions of iodine loss: study in a rat model plus in vitro analysis, Clin. Orthop. Relat. Res. (2021), doi:10.1097/CORR.0000000000001753.
[107] D. Inoue, T. Kabata, Y. Kajino, T. Shirai, H. Tsuchiya, Iodine-supported titanium implants have good antimicrobial attachment effects, J. Orthop. Sci. Off. J. Japanese Orthop. Assoc. 24 (2019) 548–551, doi:10.1016/j.jos.2018.10.010.
[108] H. Tsuchiya, T. Shirai, H. Nishida, H. Murakami, T. Kabata, N. Yamamoto, K. Watanabe, J. Nakase, Innovative antimicrobial coating of titanium implants with iodine, J. Orthop. Sci. Off. J. Japanese Orthop. Assoc. 17 (2012) 595–604, doi:10.1007/s00776-012-0247-3.
[109] S. Michelle, W. Steve, W. Thomas, Efficacy and mechanism of selenium nanoparticles as antibacterial agents, Front. Bioeng. Biotechnol. 4 (2016).
[110] P.A. Tran, N. O'Brien-Simpson, J.A. Palmer, N. Bock, E.C. Reynolds, T.J. Webster, A. Deva, W.A. Morrison, A.J. O'Connor, Selenium nanoparticles as anti-infective implant coatings for trauma orthopedics against methicillin-resistant Staphylococcus aureus and epidermidis: in vitro and in vivo assessment, Int. J. Nanomed. 14 (2019) 4613–4624, doi:10.2147/IJN.S197737.
[111] J. Holinka, M. Pilz, B. Kubista, E. Presterl, R. Windhager, Effects of selenium coating of orthopaedic implant surfaces on bacterial adherence and osteoblastic cell growth, Bone Joint J. 95-B (2013) 678–682, doi:10.1302/0301-620X.95B5.31216.
[112] N. Barraud, M.J. Kelso, S.A. Rice, S. Kjelleberg, Nitric oxide: a key mediator of biofilm dispersal with applications in infectious diseases, Curr. Pharm. Des. 21 (2015) 31–42, doi:10.2174/1381612820666140905112822.
[113] M. Li, J. Aveyard, G. Fleming, J.M. Curran, F. McBride, R. Raval, R.A. D'Sa, Nitric oxide releasing titanium surfaces for antimicrobial bone-integrating orthopedic implants, ACS Appl. Mater. Interfaces 12 (2020) 22433–22443, doi:10.1021/acsami.0c00871.
[114] E.S. DeJong, T.M. DeBerardino, D.E. Brooks, B.J. Nelson, A.A. Campbell, C.R. Bottoni, A.E. Pusateri, R.S. Walton, C.H. Guymon, A.T. McManus, Antimicrobial efficacy of external fixator pins coated with a lipid stabilized hydroxyapatite/chlorhexidine complex to prevent pin tract infection in a goat model, J. Trauma 50 (2001) 1008–1014, doi:10.1097/00005373-200106000-00006.
[115] S.M. Shiels, M. Bouchard, H. Wang, J.C. Wenke, Chlorhexidine-releasing implant coating on intramedullary nail reduces infection in a rat model, Eur. Cell. Mater. 35 (2018) 178–194, doi:10.22203/eCM.v035a13.
[116] C.A. Soriano-Souza, A.L. Rossi, E. Mavropoulos, M.A. Hausen, M.N. Tanaka, M.D. Calasans-Maia, J.M. Granjeiro, M.H.M. Rocha-Leão, A.M. Rossi, Chlorhexidine-loaded hydroxyapatite microspheres as an antimicrobial delivery system and its effect on in vivo osteo-conductive properties, J. Mater. Sci. Mater. Med. 26 (2015) 166, doi:10.1007/s10856-015-5505-4.
[117] D. Neut, R.J. Dijkstra, J.I. Thompson, C. Kavanagh, H.C. van der Mei, H.J. Busscher, A biodegradable gentamicin-hydroxyapatite-coating for infection prophylaxis in cementless hip prostheses, Eur. Cell. Mater. 29 (2015) 42–46, doi:10.22203/ecm.v029a04.
[118] C. Flores, S. Degoutin, F. Chai, G. Raoul, J.-C. Hornez, B. Martel, J. Siepmann, J. Ferri, N. Blanchemain, Gentamicin-loaded poly(lactic-co-glycolic acid) mi-

croparticles for the prevention of maxillofacial and orthopedic implant infections, Mater. Sci. Eng. C. Mater. Biol. Appl. 64 (2016) 108–116, doi:10.1016/j.msec.2016.03.064.
[119] H. Vester, B. Wildemann, G. Schmidmaier, U. Stöckle, M. Lucke, Gentamycin delivered from a PDLLA coating of metallic implants: In vivo and in vitro characterisation for local prophylaxis of implant-related osteomyelitis, Injury 41 (2010) 1053–1059, doi:10.1016/j.injury.2010.05.010.
[120] Y. Chang, C.-L. Tai, P.-H. Hsieh, S.W.N. Ueng, Gentamicin in bone cement: A potentially more effective prophylactic measure of infectionin joint arthroplasty, Bone Joint Res. 2 (2013) 220–226, doi:10.1302/2046-3758.210.2000188.
[121] P.-H. Hsieh, C.-L. Tai, P.-C. Lee, Y.-H. Chang, Liquid gentamicin and vancomycin in bone cement: a potentially more cost-effective regimen, J. Arthroplasty 24 (2009) 125–130, doi:10.1016/j.arth.2008.01.131.
[122] Y. Fillingham, A.S. Greenwald, J. Greiner, S. Oshkukov, A. Parsa, A. Porteous, M.W. Squire, Hip and knee section, prevention, local antimicrobials: proceedings of international consensus on orthopedic infections, J. Arthroplasty 34 (2019) S289–S292, doi:10.1016/j.arth.2018.09.013.
[123] K.V Holmberg, M. Abdolhosseini, Y. Li, X. Chen, S.-U. Gorr, C. Aparicio, Bio-inspired stable antimicrobial peptide coatings for dental applications, Acta Biomater. 9 (2013) 8224–8231, doi:10.1016/j.actbio.2013.06.017.
[124] A. Shukla, K.E. Fleming, H.F. Chuang, T.M. Chau, C.R. Loose, G.N. Stephanopoulos, P.T. Hammond, Controlling the release of peptide antimicrobial agents from surfaces, Biomaterials 31 (2010) 2348–2357, doi:10.1016/j.biomaterials.2009.11.082.
[125] A. Górski, E. Jończyk-Matysiak, M. Łusiak-Szelachowska, R. Międzybrodzki, B. Weber-Dąbrowska, J. Borysowski, The potential of phage therapy in sepsis, Front. Immunol. 8 (2017) 1783, doi:10.3389/fimmu.2017.01783.
[126] S.V Pavlukhina, J.B. Kaplan, L. Xu, W. Chang, X. Yu, S. Madhyastha, N. Yakandawala, A. Mentbayeva, B. Khan, S.A. Sukhishvili, Noneluting enzymatic antibiofilm coatings, ACS Appl. Mater. Interfaces 4 (2012) 4708–4716, doi:10.1021/am3010847.
[127] J.B. Kaplan, Biofilm matrix-degrading enzymes, in: G. Donelli (Ed.), Microb. Biofilms Methods Protoc., Springer New York, , New York, NY, 2014, pp. 203–213, doi:10.1007/978-1-4939-0467-9_14.
[128] J.J.T.M. Swartjes, T. Das, S. Sharifi, G. Subbiahdoss, P.K. Sharma, B.P. Krom, H.J. Busscher, H.C. van der Mei, A functional DNase I coating to prevent adhesion of bacteria and the formation of biofilm, Adv. Funct. Mater. 23 (2013) 2843–2849, doi:10.1002/adfm.201202927.
[129] M. Hentzer, M. Givskov, Pharmacological inhibition of quorum sensing for the treatment of chronic bacterial infections, J. Clin. Invest. 112 (2003) 1300–1307, doi:10.1172/JCI20074.
[130] B. Ozcelik, K.K.K. Ho, V. Glattauer, M. Willcox, N. Kumar, H. Thissen, Poly(ethylene glycol)-based coatings combining low-biofouling and quorum-sensing inhibiting properties to reduce bacterial colonization, ACS Biomater. Sci. Eng. 3 (2017) 78–87, doi:10.1021/acsbiomaterials.6b00579.
[131] B. Rémy, S. Mion, L. Plener, M. Elias, E. Chabrière, D. Daudé, Interference in bacterial quorum sensing: a biopharmaceutical perspective, Front. Pharmacol. 9 (2018) 203, doi:10.3389/fphar.2018.00203.
[132] J. Bruenke, I. Roschke, S. Agarwal, T. Riemann, A. Greiner, Quantitative comparison of the antimicrobial efficiency of leaching versus nonleaching polymer materials, Macromol. Biosci. 16 (2016) 647–654, doi:10.1002/mabi.201500266.
[133] S. Wu, J. Xu, L. Zou, S. Luo, R. Yao, B. Zheng, G. Liang, D. Wu, Y. Li, Long-lasting renewable antibacterial porous polymeric coatings enable titanium biomaterials to prevent and treat peri-implant infection, Nat. Commun. 12 (2021) 3303, doi:10.1038/s41467-021-23069-0.
[134] H. Haase, L. Jordan, L. Keitel, C. Keil, B. Mahltig, Comparison of methods for determining the effectiveness of antibacterial functionalized textiles, PLoS One 12 (2017) e0188304, doi:10.1371/journal.pone.0188304.
[135] A.J. Cunliffe, P.D. Askew, I. Stephan, G. Iredale, P. Cosemans, L.M. Simmons, J. Verran, J. Redfern, How do we determine the efficacy of an antibacterial surface? a review of standardised antibacterial material testing methods, Antibiot. (Basel, Switzerland) 10 (2021), doi:10.3390/antibiotics10091069.
[136] J. Hardes, C. von Eiff, A. Streitbuerger, M. Balke, T. Budny, M.P. Henrichs, G. Hauschild, H. Ahrens, Reduction of periprosthetic infection with silver-coated megaprostheses in patients with bone sarcoma, J. Surg. Oncol. 101 (2010) 389–395, doi:10.1002/jso.21498.
[137] G. Scoccianti, F. Frenos, G. Beltrami, D.A. Campanacci, R. Capanna, Levels of silver ions in body fluids and clinical results in silver-coated megaprostheses after tumour, trauma or failed arthroplasty, Injury 47 (Suppl 4) (2016) S11–S16, doi:10.1016/j.injury.2016.07.042.
[138] A. Karakasli, O. Hapa, O. Akdeniz, H. Havitcioğlu, Dermal argyria: cutaneous manifestation of a megaprosthesis for distal femoral osteosarcoma, Indian J. Orthop. 48 (2014) 326–328, doi:10.4103/0019-5413.132528.
[139] M. Glehr, A. Leithner, J. Friesenbichler, W. Goessler, A. Avian, D. Andreou, W. Maurer-Ertl, R. Windhager, P.-U. Tunn, Argyria following the use of silver-coated megaprostheses: no association between the development of local argyria and elevated silver levels, Bone Joint J. 95-B (2013) 988–992, doi:10.1302/0301-620X.95B7.31124.
[140] B. Hussmann, I. Johann, M.D. Kauther, S. Landgraeber, M. Jäger, S. Lendemans, Measurement of the silver ion concentration in wound fluids after implantation of silver-coated megaprostheses: correlation with the clinical outcome, Biomed Res. Int. 2013 (2013) 763096, doi:10.1155/2013/763096.
[141] H. Wafa, R.J. Grimer, K. Reddy, L. Jeys, A. Abudu, S.R. Carter, R.M. Tillman, Retrospective evaluation of the incidence of early periprosthetic infection with silver-treated endoprostheses in high-risk patients: case-control study, Bone Joint J. 97-B (2015) 252–257, doi:10.1302/0301-620X.97B2.34554.
[142] A. Piccioli, F. Donati, G. Di Giacomo, A. Ziranu, S. Careri, M.S. Spinelli, S. Giannini, G. Giannicola, C. Perisano, G. Maccauro, Infective complications in tumour endoprostheses implanted after pathological fracture of the limbs, Injury 47 (Suppl 4) (2016) S22–S28, doi:10.1016/j.injury.2016.07.054.
[143] F. Donati, G. Di Giacomo, S. D'Adamio, A. Ziranu, S. Careri, M. Rosa, G. Maccauro, Silver-coated hip megaprosthesis in oncological limb savage surgery, Biomed Res. Int. 2016 (2016) 9079041, doi:10.1155/2016/9079041.
[144] J. Hardes, M.P. Henrichs, G. Hauschild, M. Nottrott, W. Guder, A. Streitbuerger, Silver-coated megaprosthesis of the proximal tibia in patients with sarcoma, J. Arthroplasty 32 (2017) 2208–2213, doi:10.1016/j.arth.2017.02.054.
[145] D. Zajonz, U. Birke, M. Ghanem, T. Prietzel, C. Josten, A. Roth, J.K.M. Fakler, Silver-coated modular Megaendoprostheses in salvage revision arthroplasty after periimplant infection with extensive bone loss - a pilot study of 34 patients, BMC Musculoskelet. Disord. 18 (2017) 383, doi:10.1186/s12891-017-1742-7.
[146] A. Streitbuerger, M.P. Henrichs, G. Hauschild, M. Nottrott, W. Guder, J. Hardes, Silver-coated megaprostheses in the proximal femur in patients with sarcoma, Eur. J. Orthop. Surg. Traumatol. 29 (2019) 79–85, doi:10.1007/s00590-018-2270-3.
[147] M.R. Medellin, T. Fujiwara, R. Clark, J.D. Stevenson, M. Parry, L. Jeys, Mechanisms of failure and survival of total femoral endoprosthetic replacements, Bone Joint J. 101-B (2019) 522–528, doi:10.1302/0301-620X.101B5.BJJ-2018-1106.R1.
[148] M.C. Parry, M.K. Laitinen, J.I. Albergo, C.L. Gaston, J.D. Stevenson, R.J. Grimer, L.M. Jeys, Silver-coated (Agluna®) tumour prostheses can be a protective factor against infection in high risk failure patients, Eur. J. Surg. Oncol. J. Eur. Soc. Surg. Oncol. Br. Assoc. Surg. Oncol. 45 (2019) 704–710, doi:10.1016/j.ejso.2018.12.009.
[149] A. Sambri, R. Zucchini, C. Giannini, E. Zamparini, P. Viale, D.M. Donati, M. De Paolis, Silver-coated (PorAg(®)) endoprosthesis can be protective against re-infection in the treatment of tumor prostheses infection, Eur. J. Orthop. Surg. Traumatol. 30 (2020) 1345–1353, doi:10.1007/s00590-020-02705-3.
[150] C. Zoccali, G. Scoccianti, R. Biagini, P.A. Daolio, F.L. Giardina, D.A. Campanacci, Antibacterial hydrogel coating in joint mega-prosthesis: results of a comparative series, Eur. J. Orthop. Surg. Traumatol. (2021), doi:10.1007/s00590-021-02884-7.
[151] M.A. O'Malley, The first eukaryote cell: an unfinished history of contestation, Stud. Hist. Philos. Sci. Part C Stud. Hist. Philos. Biol. Biomed. Sci. 41 (2010) 212–224, doi:10.1016/j.shpsc.2010.07.010.
[152] C.L. Ventola, The antibiotic resistance crisis: part 1: causes and threats, P T. 40 (2015) 277–283.
[153] M.P. Siljander, A.H. Sobh, K.C. Baker, E.A. Baker, L.M. Kaplan, Multidrug-resistant organisms in the setting of periprosthetic joint infection-diagnosis, prevention, and treatment, J. Arthroplasty 33 (2018) 185–194, doi:10.1016/j.arth.2017.07.045.
[154] K. Bruellhoff, J. Fiedler, M. Möller, J. Groll, R.E. Brenner, Surface coating strategies to prevent biofilm formation on implant surfaces, Int. J. Artif. Organs. 33 (2010) 646–653, doi:10.1177/039139881003300910.
[155] J. Fiedler, J. Groll, E. Engelhardt, P. Gasteier, C. Dahmen, H. Kessler, M. Moeller, R.E. Brenner, NCO-sP(EO-stat-PO) surface coatings preserve biochemical properties of RGD peptides, Int. J. Mol. Med. 27 (2011) 139–145, doi:10.3892/ijmm.2010.553.
[156] J. Groll, J. Fiedler, K. Bruellhoff, M. Moeller, R.E. Brenner, Novel surface coatings modulating eukaryotic cell adhesion and preventing implant infection, Int. J. Artif. Organs. 32 (2009) 655–662, doi:10.1177/039139880903200915.
[157] C. Neuerburg, S. Recknagel, J. Fiedler, J. Groll, M. Moeller, K. Bruellhoff, H. Reichel, A. Ignatius, R.E. Brenner, Ultrathin sP(EO-stat-PO) hydrogel coatings are biocompatible and preserve functionality of surface bound growth factors in vivo, J. Mater. Sci. Mater. Med. 24 (2013) 2417–2427, doi:10.1007/s10856-013-4984-4.
[158] S.M. Kurtz, E. Lau, H. Watson, J.K. Schmier, J. Parvizi, Economic burden of periprosthetic joint infection in the United States, J. Arthroplasty 27 (2012) 61–65 e1, doi:10.1016/j.arth.2012.02.022.
[159] K. Garfield, S. Noble, E. Lenguerrand, M.R. Whitehouse, A. Sayers, M.R. Reed, A.W. Blom, What are the inpatient and day case costs following primary total hip replacement of patients treated for prosthetic joint infection: a matched cohort study using linked data from the National Joint Registry and Hospital Episode Statistics, BMC Med. 18 (2020) 335, doi:10.1186/s12916-020-01803-7.
[160] T. Shirai, H. Tsuchiya, R. Terauchi, S. Tsuchida, N. Mizoshiri, Y. Mori, A. Takeuchi, K. Hayashi, N. Yamamoto, K. Ikoma, T. Kubo, A retrospective study of antibacterial iodine-coated implants for postoperative infection, Medicine (Baltimore) 98 (2019) https://journals.lww.com/md-journal/Fulltext/2019/11080/A_retrospective_study_of_antibacterial.72.aspx.
[161] T. Kabata, T. Maeda, Y. Kajino, K. Hasegawa, D. Inoue, T. Yamamoto, T. Takagi, T. Ohmori, H. Tsuchiya, Iodine-supported hip implants: short term clinical results, Biomed Res. Int. 2015 (2015) 368124, doi:10.1155/2015/368124.