International Journal of Medical Microbiology 309 (2019) 1–12

Contents lists available at ScienceDirect

International Journal of Medical Microbiology

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

An update on recent developments in the prevention and treatment of *Staphylococcus aureus* biofilms

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ARTICLE INFO

Keywords:
Staphylococcus aureus
Biofilms
Antibiotics
Nanoparticles
Phytochemicals
Phages

ABSTRACT

Staphylococcus aureus (*S. aureus*) readily forms biofilms on prosthetic devices such as the pacemakers, heart valves, orthopaedic implants, and indwelling catheters. Its biofilms are recalcitrant to antibiotic therapy and pose a serious burden in the healthcare setting as they drastically increase the treatment cost and morbidity of the patient. Prevention and treatment of staphylococcal biofilms has therefore been an area of active research for the past two decades. While catheters coated with different antiseptics and antibiotics capable of preventing *S. aureus* biofilm formation have been developed, an effective therapy for the dispersal and treatment of established staphylococcal biofilms is not yet available. Hence, many studies have focused on developing novel therapeutic strategies that can tackle established *S. aureus* biofilm associated infections. This has led to the identification of different phytochemicals (*e.g.*, tannic acid, ellagic acid, xanthohumol *etc*), enzymes (*e.g.*, Dnases, lysostaphin, α-amylase, hyaluronidase and proteases *etc.*), sulfahydrl compounds (*e.g.*, dithiothreitol, 2-mercaptoethanol), nanoparticles (*e.g.*, gold, silver, iron, copper and selenium), phage cocktails, antibodies and metal chelators. Apart from the conventional techniques, the therapeutic effects of ultra sound, shock waves and photodynamic therapy for treating *S. aureus* biofilms are also being investigated. Clinical validation of these studies will equip the medical field with alternate preventive and treatment methods against staphylococcal biofilm infections. This review provides recent updates on the preventive and therapeutic strategies explored to eradicate staphylococcal biofilm formation and related infections.

1. Introduction

Staphylococcus aureus (*S. aureus*), is one of the leading human pathogens that is responsible for a number of community acquired, nosocomial and biofilm related infections worldwide (Archer et al., 2011a, 2011b; Tong et al., 2015). Its pathogenic efficiency can be attributed to wide range of virulence factors it produces to thwart host immune responses (De Leo et al., 2009; Thammavongsa et al., 2015). *S. aureus* infections, particularly its biofilm related infections are recalcitrant to clearance by antimicrobials and host defence molecules. Biofilms also increase the opportunity for the transfer the resistance genes between bacteria (Archer et al., 2011a, 2011b; Donlan, 2002; Flemming et al., 2016; Hall-Stoodley et al., 2004; Römling and Balsalobre, 2012; Weigel et al., 2007). In clinical scenario *S. aureus* forms biofilms on the surfaces of the catheters (intravenous catheters, urinary catheters, dialysis catheters *etc.*) and implanted medical devices (fluid shunts, joint prostheses and pacemakers *etc*). *S. aureus* in the biofilms are resistant to antibiotics and innate host defence mechanisms, such as antimicrobial peptides and neutrophil phagocytosis (Otto, 2008). Treatment in such cases requires the replacement of the infected catheters and implants with new ones and subsequent treatment with antibiotics resulting in drastic increase in the treatment cost (Lebeaux et al., 2014). On-going research in the last decades identified a number of prevention and treatment strategies for tackling *S. aureus* biofilm infections. The updates regarding the preventive and therapeutic strategies explored recently to eradicate staphylococcal biofilm formations are discussed in this review.

2. *S. aureus* biofilm structure

Biofilms are organized assemblage of sessile bacterial cells protected by an extracellular matrix (ECM). *S. aureus* forms biofilm on catheters and other biomaterials surfaces. Previous studies have demonstrated that soon after implantation, catheters and other biomaterials get coated with plasma and host ECM proteins such as fibrinogen, fibronectin, thrombospondin and vitronectin (Lundberg et al., 1997). *S.*

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https://doi.org/10.1016/j.ijmm.2018.11.002
Received 31 March 2018; Received in revised form 19 November 2018; Accepted 26 November 2018

aureus has tremendous capacity to bind to both uncoated, as well as host protein coated catheters and other biomaterials. *S. aureus* biofilm development proceeds in three steps: attachment, accumulation and dispersal (Boles and Horswill, 2008; Chessa et al., 2016; Joo and Otto, 2012; Laverty et al., 2013; Le et al., 2014; Lister and Horswill, 2014; Moormeier et al., 2014). The biofilm formation begins with the initial attachment of *S. aureus* cells to the uncoated or plasma and host ECM proteins coated catheter and biomaterial surfaces, followed by formation of bacterial aggregates (Otto, 2008). Attachment to the surface(s) is mediated by hydrophobic, electrostatic and *van der waals* interactions (Renner and Weibel, 2011). The development of multi-layered structure of biofilm requires intercellular aggregation of the proliferating cells through the production of ECM (Otto, 2008). In general, ECM in *S. aureus* is composed of polysaccharides, teichoic acids, extracellular DNA (eDNA) and several surface proteins of *S. aureus* (Schwartz et al., 2016), however the amount of individual components present within the biofilms varies among the different *S. aureus* isolates (Sugimoto et al., 2018). The polysaccharide in staphylococcal biofilm is named as polysaccharide intracellular adhesion (PIA). PIA is also known as polymeric *N*-acetyl- glucosamine (PNAG) and both abbreviations are used to name actually the same molecule (Arciola et al., 2015; Lister and Horswill, 2014). PIA/ PNAG is composed of β-1, 6-linked *N*-acetyl glucosamine residues (80–85%) and an anionic fraction with a lower content of non-*N*-acetylated D-glucosaminyl residues that contain phosphate and ester-linked succinate (15–20%) (Archer et al., 2011a, 2011b) with a net positive charge that promotes intercellular aggregation and attachment of *S. aureus* to inert surfaces (Shirtliff et al., 2002). PIA/ PNAG synthesis in this bacterium is mediated by the i*caADBC* operon (Cramton et al., 1999; Fluckiger et al., 2005). Staphylococcal surface proteins such as autolysin AtlA, plays an important role in stabilizing the biofilm structure by inducing eDNA release (Bose et al., 2012; Speziale et al., 2014). The released eDNA will be held within the ECM by secreted bacterial proteins such as integration host factor (IHF) and histone-like (HU) proteins that belong to the DNABII family. Other *S. aureus* surface proteins like Protein A (Merino et al., 2009; Toledo-Arana et al., 2005), fibronectin and fibrinogen binding factors (FnBPA and FnBPB, ClfA and ClfB) (Fitzpatrick et al., 2005; O'Neill et al., 2009, 2008; Vergara-Irigaray et al., 2009), serine aspartate repeat protein (SdrC) (Barbu et al., 2014), surface associated staphylococcal protein G (SasG) (Corrigan et al., 2007), biofilm associated protein (Bap) (Cucarella et al., 2004) and accumulation associated proteins (Aap) (Schaeffer et al., 2015) contribute to biofilm matrix development and stabilization (Lister and Horswill, 2014) (Fig. 1). *S. aureus* surface proteins Atl, FnBPA and FnBPB, ClfA, ClfB and teichoic acids play vital role in the initial attachment of *S. aureus* to the host protein coated surfaces.

Sugimoto et al., studied the importance of each component of ECM (eDNA, proteins and PIA/ PNAG) in biofilm formation among the different *S. aureus* clinical strains using DNase I, Proteinase K and Dispersin B. DNase I was found to be most effective in biofilm dispersion, followed by Proteinase K. The biofilm dispersion activity of Dispersin B, which cleaves PIA/ PNAG, was restricted only to *S. aureus* strains that produce PIA/ PNAG. Dispersin also showed significant inverse correlations with that of Proteinase K, suggesting independent roles of PIA/ PNAG and proteins in each biofilm. ECM profiling of different clinical strains revealed that while eDNA was present in all strains, PIA/ PNAG was present only in a small number of isolates (Sugimoto et al., 2018).

3. Therapeutic strategies to prevent biofilm formation and disruption

The increasing incidence of biofilm associated *S. aureus* infections necessitates the development of novel methods to treat them. The main problem associated with staphylococcal biofilms is their resistance towards the antibiotics and host defence mechanisms. Biofilm forming property endows the pathogenic strains with increased resistance towards antibiotics and host defence factors (De Leo et al., 2009; Günther et al., 2009; Thurlow et al., 2011). This resistance is predominantly effectuated through the diffusion barrier action of the polysaccharide matrix (Singh et al., 2016). Additionally, the anoxic conditions and lack of sufficient nutrients within the biofilm force *S. aureus* cells into a dormant state with down regulated metabolism and impaired cell division leading to a population of slow growing persistent *S. aureus* cells which impede the anti-staphylococcal activity of most antibiotics (Chung and Toh, 2014; Lebeaux et al., 2014; Lewis, 2008; Wood et al., 2013).

Tackling the antibiotic resistance of *S. aureus* in biofilms requires development of suitable treatment(s) options. The efforts that were made for the prevention and treatment of *S. aureus* biofilms can broadly be categorized into the following major groups: (i) use of existing antibiotics in combinations; (ii) repurposing of existing drugs; (ii) identification of new anti-biofilm agents; (iii) use of laser Shock waves (LSW), ultra sound (US) and photodynamic therapies (PDT); and (iv) development of medical implants and catheters with modified surfaces that hinder the *S. aureus* attachment and biofilm development. This review provides a concise overview of the recent findings in each of the above mentioned topics (Fig. 2).

3.1. Antibiotics and their combinations

Several approaches were considered to use antibiotics in an appropriate way to tackle *S. aureus* biofilm infections, which included both treatment and prevention strategies. Treatment strategies involved exploring the use of antibiotics at higher concentrations or in different combinations to achieve maximum therapeutic effects. As *S. aureus* biofilms are resistant to antibiotics at their minimum inhibitory concentrations (MIC) it became a necessity to determine the exact minimum biofilm inhibition concentrations (MBIC; the minimum concentration that prevents biofilm formation) and biofilm eradication concentrations (MBEC; minimum concentration necessary to eradicate preformed biofilm) of different antibiotics (Kotulová and Slobodníková, 2010; Manner et al., 2017; Mottola et al., 2016). The MIC, MBIC and MBEC values of different antibiotics against *S. aureus* were determined and presented in several previously published articles (LaPlante and Mermel, 2009; Mottola et al., 2016; Saginur et al., 2006). In all cases the MBIC and MBEC values of antibiotics were much higher than their respective MICs. For example, the MIC, MBIC and MBEC values of vancomycin against *S. aureus* were 1–2, 16 (8–16 times more than MIC) and > 256 μg/ml (at least 16 times more than MBIC); and the MIC and MBIC values of linezolid against *S. aureus* was 2 and > 64 μg/ml. The higher MBIC and MBEC values make it difficult to use most antibiotics to treat *S. aureus* biofilm infections, as at MBIC and MBEC values these antibiotics may exert cellular toxicity. As single antibiotics in general were not effective in eradicating *S. aureus* biofilms at their MICs, efforts were made to identify suitable antibiotic combinations that can disperse biofilm at their MICs. *In vitro* studies identified several antibiotic combinations that can disperse *S. aureus* biofilms at their MICs. The combinations that were found to be effective against methicillin sensitive *S. aureus* are oxacillin (OXA), azithromycin (AZM), fusidic acid (FA); OXA, gentamicin (GEN), FA; rifampicin (RIF), cefazolin (CFZ); RIF, vancomycin (VAN), GEN; CFZ, AZM, FA; CFZ, ciprofloxacin (CIP), FA; VAN, FA; VAN, AZM, FA; GEN, FA; AZM, CIP, FA; RIF, VAN; while combinations of RIF, VAN, FA and RIF, CIP, FA were bactericidal against methicillin resistant *S. aureus* biofilms (Saginur et al., 2006). Rifampin was found to be the single most active antibiotic agent against *S. aureus* biofilms (Kadurugamuwa et al., 2003; Kotulová and Slobodníková, 2010).

Implants and catheters coated with different antiseptics (silver sulfadiazine, chlorhexidine *etc*) and antibiotics (minocycline, rifampin *etc*) were developed as strategies to prevent *S. aureus* foreign body associated biofilm development. Sampath et al have compared the biofilm

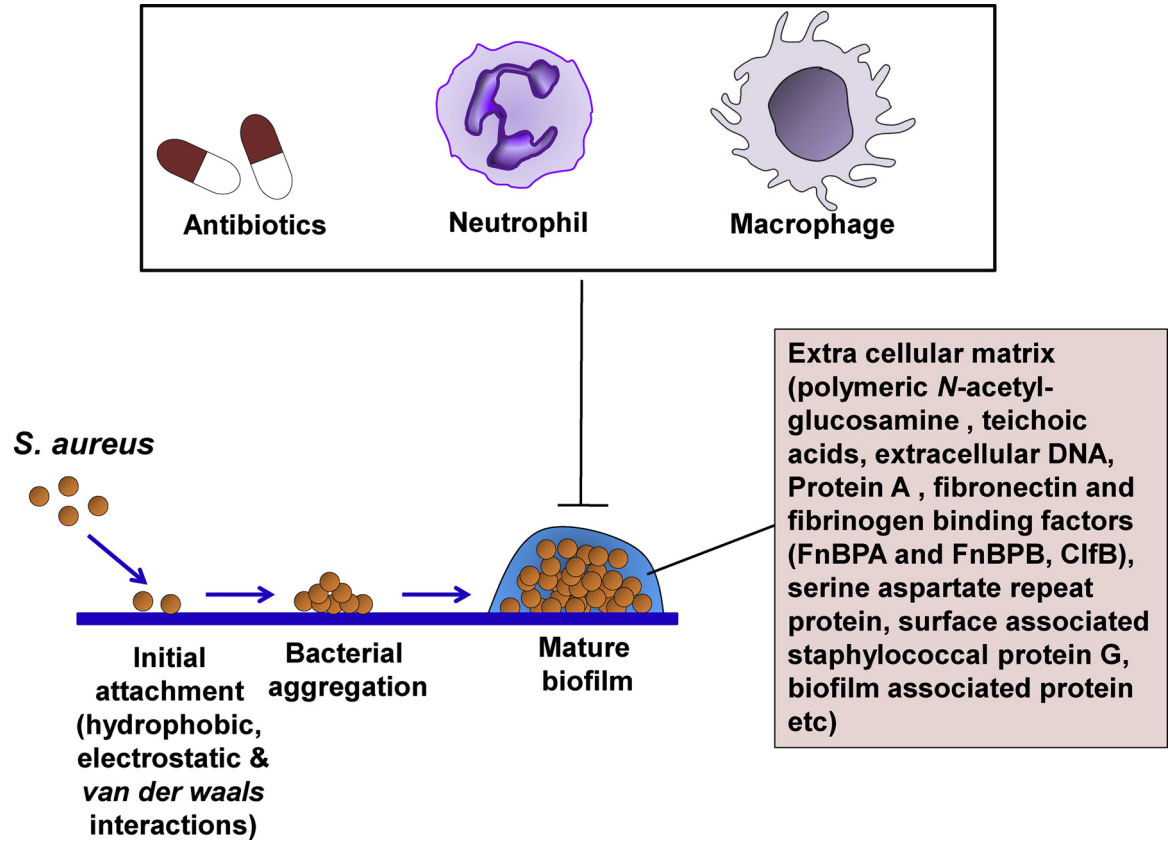

Fig. 1. Steps involved in *S. aureus* biofilm development and its resistance mechanisms. Biofilms form by initial attachment of *S. aureus* to biotic or abiotic surfaces. Subsequently, *S. aureus* divides to form multilayer aggregates and later mature biofilm, where *S. aureus* get encased into the ECM composed of polymeric *N*-acetyl glucosamine, teichoic acids, and proteins. Biofilm embedded *S. aureus* are resistant to antibiotics and innate host defence mechanisms, such as neutrophil and macrophage medicated phagocytosis.

inhibitory properties of catheters containing silver sulfadiazine and chlorhexidine on the external surface and chlorhexidine in the lumens to the catheters impregnated with minocycline on its external surfaces and rifampin on its luminal surfaces in a rat model. Both catheters prevented *S. aureus*biofilm development (Sampath et al., 2001). Using a murine model of bacterial colonization of subcutaneously inserted catheters, Tran et al, demonstrated that organoselenium antimicrobial agent selenocyanatodiacetic acid (SCAA) coated hemodialysis catheters (HDC) inhibit *S. aureus* biofilm formation (Tran et al., 2012). A major concern of the antibiotic coated implants and catheters is that their use might lead to the development of resistant strains.

3.2. Identification of new anti-biofilm agents

Constant efforts were also made to develop new anti-biofilm agents which led to the identification of several repurposed drugs, phytochemicals, metal chelators, nanoparticles, antimicrobial peptides (AMPs), enzymes, bacteriophages and antibodies that can combat staphylococcal biofilm associated infections. The next section focuses on the potential of these agents.

3.2.1. Repurposed drugs

Development of new therapeutic molecules requires extensive research with costs running into hundreds of millions of dollars over longer duration of time (usually 10–12 years). As development of completely new and effective drug molecules is increasingly becoming difficult, alternative strategies such as repurposing of existing Food and Drug Administration (FDA) approved drugs for the treatment of multiple diseases are gaining popularity. This approach has several advantages, including avoidance of high costs and long duration of time required for development of novel antibiotics, also there is an added benefit of utilizing compounds with known pharmacological properties (Deftereos et al., 2011; Corsello et al., 2017; Hernandez et al., 2017). For example, Auranofin, a drug used for the treatment of rheumatoid arthritis, has been repurposed to treat *S. aureus*, *K. pneumoniae*, HIV and few parasitic infections (Caroli et al., 2012; Chirullo et al., 2013; Sharlow et al., 2014; Aguinagalde et al., 2015). Enoxacin, a broad-spectrum fluoroquinolone antibacterial agent has been repurposed to treat fungal infections (Breger et al., 2007). Active research in drug repurposing led to the identification of a number of existing drug molecules such as thioridazine, niclosamide, carmofur and auranofin with anti-biofilm activities. Thioridazine, an anti-psychotic medicine used to treat schizophrenia potentiated the activity of tobramycin, linezolid and flucloxacillin against *S. aureus* biofilm cells (Van den Driessche et al., 2017). Niclosamide, a medication used to treat tapeworm infections; Carmofuran, an antineoplastic agent and auranofin, an anti-rheumatic agent were also found to act against pre-formed staphylococcal biofilms (Gwisai et al., 2017; Thangamani et al., 2016; Torres et al., 2016; Van den Driessche et al., 2017). However, the anti-biofilm activity of all the above-mentioned repurposed drugs was tested only under *in vitro* conditions and requires further *in vivo* validation.

3.2.2. Chelators and sulfhydryl compounds

Cations (*e.g.*, Mg^{2+}, Fe^{2+}, Ca^{2+}) play important role in microbial adherence, biofilm formation, and bacterial growth by promoting inter bacterial interactions, aggregation and inhibition of PIA/ PNAG synthesis. High affinity metal ion chelators such as ethylenediamine tetraacetic acid (EDTA), ethylene glycol tetra acetic acid (EGTA) and tri-sodium citrate (TSC) inhibit biofilm formation as they sequester the ions and obstruct inter bacterial interactions (Abraham et al., 2012).

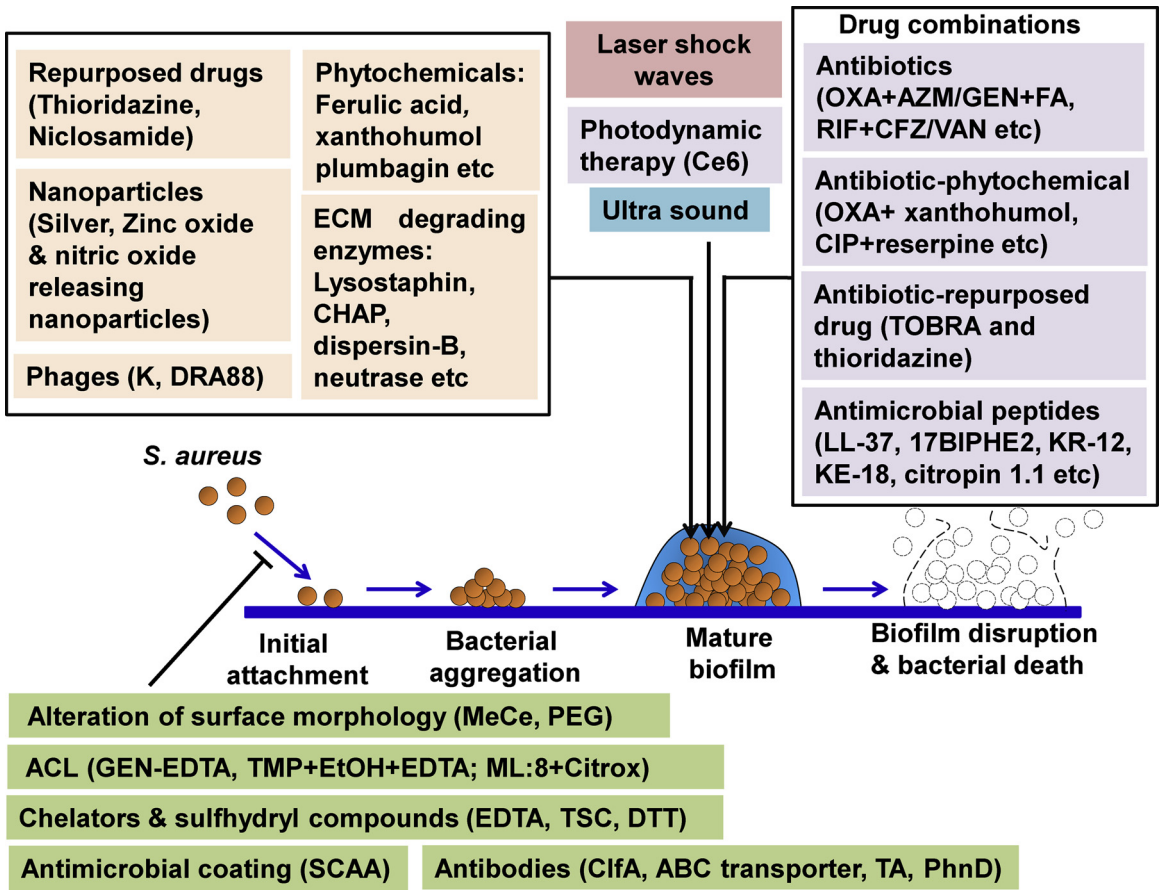

Fig. 2. Recent developments in prevention and treatment strategies against *S. aureus* biofilm. Biofilm formation could be inhibited using implants with altered surface morphology and anti-microbial coating. Chelators and sulfhydryl compounds have also demonstrated to possess biofilm inhibitory activities. A number of compounds and their combinations (eg, repurposed drugs, nanoparticles, antibodies, phages, enzymes *etc*) are also identified to have activities against mature biofilm. Abbreviations: ECM = extracellular matrix; CHAP = Cysteine, histidine dependant amidohydrolase/peptidase; Ce6 = chlorin e6; OXA = oxacillin; AZM = azithromycin; GEN = gentamicin; FA = fusidic acid; RIF = rifampicin; CFZ = cefazolin; VAN = vancomycin; CIP = ciprofloxacin; TOBRA = tobramycin; TMP = trimethoprim; EtOH = ethanol; EDTA = ethylenediamine tetraacetic acid; TSC = tri-sodium citrate; DTT = dithiothreitol; SCAA = selenocyanatodiacetic acid.

It was observed that sulfhydryl compounds such as dithiothreitol (DTT), beta-mercaptoethanol and cysteine also cause significant inhibition of biofilm formation in *S. aureus*. They mainly affect the initial adhesion of *S. aureus*. Thiol treatment upregulates expression of several proteins involved in the Embden Meyerhof pathway and Pentose Phosphate pathway that boost glucose metabolism resulting in reduced Uridinediphosphate*N*-acetylglucosamine (UDP-GlcNAc) availability for PIA/ PNAG synthesis. As UDP-GlcNAc is required for PIA/ PNAG synthesis (Arciola et al., 2015; Wu et al., 2011) and synthesis of PIA/ PNAG is necessary for *S. aureus* biofilm development, decreased availability of UDP-GlcNAc leads to a reduction in its biofilm development.

3.2.3. Nanoparticles

Nanotechnology is gaining tremendous importance in recent days as drug delivery vehicles. The success of nanoparticles as therapeutic agents is attributed to their small size (less than 1 μm) and higher surface to mass ratio which facilitate effective interactions with the biological systems (Singh and Lillard, 2009). The field of nanotechnology provides novel approaches to tackle *S. aureus* biofilm associated infections. Several nanomaterials, nanoparticles (NP) and drug encapsulated nanoparticles have been shown to possess better antibacterial and anti-biofilm activities. The anti-biofilm activities of nanoparticles are due to their enhanced interaction and penetrative capabilities within the biofilm matrix than the existing free drug molecules (Mu et al., 2016; Pelgrift and Friedman, 2013). Silver and Zinc oxide (ZnO) nanoparticles; and sustained nitric oxide releasing nanoparticles were found to interfere with *S. aureus* adhesion and thus prevent its biofilm formation (Mihu et al., 2017). Additionally, synergistic anti-biofilm activity of silver and Zinc oxide (ZnO) nanoparticles was observed when used in combinationwith antibiotics (Chaudhari et al., 2012). Chaudhari et al., demonstrated that while gentamicin and chloramphenicol failed to disperse *S. aureus* biofilms individually, synergistic effect of silver nanoparticles along with these antibiotics resulted in enhanced dispersion of *S. aureus* biofilms (Chaudhari et al., 2012). Applerot et al., demonstrated synergism between chloramphenicol, ampicillin, erythromycin and ZnO nanoparticles against *S. aureus* biofilm (Applerot et al., 2012). Lellouche et al., reported the antimicrobial activity of fluoride nanomaterials (Lellouche et al., 2009). Magnesium fluoride (MgF_2) NP and yttrium fluoride (YF_3) NP coated catheters were able to reduce *S. aureus* colonization on catheter surfaces significantly compared to the uncoated controls (Lellouche et al., 2012b, 2012a, 2009). Phosphatidylcholine-decorated gentamicin loaded gold nanoparticles also demonstrated superior anti-biofilm efficacy against *S. aureus* (Mu et al., 2016).

3.2.4. Antimicrobial peptides (AMPs)

Anti-microbial peptides (AMPs) are key components of innate immunity in humans and other higher organisms. Most AMPs are positively charged, amphipathic in nature; and shorter than 50 amino acids. AMPs can bind to and disrupt bacterial membranes (Diamond et al., 2009; Li et al., 2017). Till date, more than 5000 AMPs have been discovered or synthesized (Zhao et al., 2013). In addition to their

antimicrobial activity some of these AMPs also possess anti-biofilm activity against *S. aureus*. These include RNAIII-inhibiting peptide (RIP; YSPWTNF) (Giacometti et al., 2003) and LL-37 (LLGDFFRKSKEKIGK-EFKRIVQRIKDFLRNLVPRTES). However, when tested under *in vivo* conditions RIP exhibited low anti-biofilm activity (Zhou et al., 2016) and LL-37 was not very stable (Moncla et al., 2011). Thus, several of their derivatives were synthesized to enhance their *in vivo* antimicrobial and anti-biofilm activities. The derivatives which showed enhanced anti-biofilm activities include synthetic RIP (16P-AC; $_{CH3CO}$-YKPVTNF-ST-YKPVTNF-$_{CONH2}$) (Zhou et al., 2016) and D-LL-37 (protease-resistant enantiomer of LL-37) (Dean et al., 2011). Several other truncated derivatives of LL-37 were also synthesized, *viz*., GF-17 (FKRIVQRIKDFLRNLV; residues 17 to 32 of LL-37 and protease sensitive), 17BIPHE2 (GXKRIVQRIKDXLRNLV, where X is biphenylalanine; was developed based on GF-17 sequence, was protease resistant and was found to be more effective than LL-37 and GF-17) (Mishra et al., 2015), KE-18 (KEFKRIVQRIK; residues 15 to 25 of LL-37) and KR-12 (KRIVQRIKDFLR; residues 18 to 29 of LL-37) (Luo et al., 2017). NA-CATH:ATRA1-ATRA1 (KRFKKFFKKLKNSVKKRFKKFFKKLKVIGVTFPF), a derivative of natural snake cathelicidin also demonstrated *in vitro* anti-biofilm activity against *S. aureus* (Dean et al., 2011). Other AMPs such as Bac8-c^{2,5Leu} (RLWVLWRR; synthetic) (Zapotoczna et al., 2017), tachyplesin I (KWCFRVCYRGICYRRCR; derived from horseshoe crab) (Jorge et al., 2017), citropin 1.1 (GLFDVIKKVASVIGGL; green tree frog *Litoria citropa*), temporin A (FLPLIGRVLSGIL; derived from the frog *Rana temporaria*) (Dawgul et al., 2016) and Larvae Immune Peptides 7 complex (FLIP7; isolated from the maggots of blowfly *Calliphora vicina*) (Gordya et al., 2017) demonstrated anti-biofilm properties against *S. aureus*. Although larger than AMPs, human Short-palate lung and nasal epithelial clone 1 (SPLUNC1) protein possess anti-biofilm activity against S. aureus. SPLUNC1 is a 256-amino acid multifunctional protein secreted by the human respiratory tract. SPLUNC1 helps in maintaining fluid homeostasis in airway epithelia and possess antimicrobial activity (Britto and Cohn, 2015). Yu et al., have demonstrated that the α4 region (ILKPGGGTSGGLLGGLLGKVTSVIPGLNNI) of SPLUNC1 is responsible for its anti-biofilm activity. A synthetic derivative of α4 region, which was named as α4M1 peptide (ILKKWWGTSGGLLGGLLGKVTSVIKGLNNI) demonstrated at least seven fold higher *in vitro* anti-biofilm activity than α4 region (Yu et al., 2018). Among these AMPs only the activity of RIP, 16P-AC and Bac8-c^{2,5Leu} were validated both under *in vitro* and *in vivo* conditions. 16P-AC and Bac8c^{2,5Leu} demonstrated significant biofilm disrupting activity, whereas RIP showed superior biofilm disrupting activity only in combination with antibiotics (Table 1).

3.2.5. Enzymes as biofilm disrupting agents

Research is underway to understand the efficacy of different enzymes that can destabilize *S. aureus* biofilms through degradation of its cell wall or ECM. Different cell wall hydrolases that can degrade the pentaglycine bridges in the *S. aureus* cell wall (eg, Lysostaphin, Cysteine histidine dependant amidohydrolase/peptidase, endolysins) (Bradford, 2011; Ibberson et al., 2016; Kaplan et al., 2004), proteases (eg, V8 protease, staphopains and cysteine proteases) and DNases were identified to be effective against *S. aureus* biofilms. Lysostaphin, a glycyl glycine endopeptidase, which cleaves the pentaglycine cross-bridge of staphylococcal cell wall possess the ability to disrupt *S. aureus* biofilms (Kokai-Kun et al., 2009). Bacteriophage endolysins including Cysteine, histidine dependant amidohydrolase/peptidase from phage K (CHAPK), LysH5 from phage vB-SauS-phiIPLA88 and endolysin from phi11 phage have been found to possess the ability to lyse and disrupt the complex structure of staphylococcal biofilms (Fenton et al., 2013; Gutiérrez et al., 2014; Sass and Bierbaum, 2007; Walencka et al., 2005; Wu et al., 2003).

Among the different glycogen hydrolases, α-amylase was effective in biofilm detachment, while dispersin B was found to reduce interbacterial adherence and bacteria-surface attachment of *S. aureus* (Bradford, 2011; Kaplan et al., 2004). Although the mechanism of action of α-amylase on *S. aureus* biofilm is not elucidated so far, dispersin B acts by hydrolysing the PNAG of biofilm matrix (Kaplan et al., 2004). Hyaluronidase, another glycogen hydrolase was also effective in dispersing *S. aureus* biofilm. It cleaves the glycosidic linkages of hyaluronic acid of the host extracellular matrix (ECM) used by *S. aureus* for biofilm matrix formation thus leading to biofilm disruption (Ibberson et al., 2016).

In the quest for methods that can disrupt the biofilms, attempts have also been made to use proteases encoded by staphylococci. *S. aureus* encodes 10 proteases of which seven are serine proteases [SplA-F and V8 protease (sspA)], two are cysteine proteases [Staphopain A (ScpA) and Staphopain B (SspB)] and one is a metalloprotease [Aureolysin (Aur)] (Kolar et al., 2013; Shaw et al., 2004). Among these proteases, V8 protease was found to reduce *S. aureus* biofilm formation and promote biofilm detachment through autolysin (AtlA) inactivation (Chen et al., 2013). The role of staphopains ScpA and SspB, cysteine proteases, in hampering biofilm integrity has been demonstrated by Mootz et al., (Mootz et al., 2013). While both ScpA and SspB prevent biofilm formation, ScpA has also been shown to disperse established biofilms (Mootz et al., 2013). Aureolysin has been found to proteolytically cleave and activate SspA which in turn activates SspB (Nickerson et al., 2010). Loughran et al., confirmed that Aur prevents biofilm formation and disperses preformed biofilms (Loughran et al., 2014).

Proteases such as proteinase K, trypsin, LasB elastase, flavourzyme and neutrase from external sources have also been found to be effective in biofilm disassembly (Gilan and Sivan, 2013). Proteinase K and trypsin are serine proteases that are verified as efficient inhibitors of biofilm formation (Elchinger et al., 2014; Gilan and Sivan, 2013). LasB, a staphylolytic enzyme secreted by *Pseudomonas aeruginosa* cleaves the pentaglycine bridge of *S. aureus*. Studies have shown that LasB has a mode of action similar to that of lysostaphin and both the enzymes have demonstrated the ability to prevent biofilm formation and disperse preformed *S. aureus* biofilms. Other enzymes which were found to be effective against staphylococcal biofilms were flavourzyme, an aminoprotease from *Aspergillus oryza* and neutrase from *Bacillus amyloliquefaciens*. However, flavourzyme was only effective against *S. epidermidis*, whereas neutrase was effective against both *S. aureus* and *S. epidermidis*. The reason behind the specific anti-biofilm activity of flavourzyme against *S. epidermidis* and not against *S. aureus* is not yet studied (Elchinger et al., 2014).

Similar to proteases, nucleases from different sources also showed anti-biofilm efficacy. Mann et al and Kiedrowski et al., reported the anti-biofilm activity of staphylococcal nuclease (Nuc) (Kiedrowski et al., 2011; Mann et al., 2009). The recombinant human DNase (DnaseI) was also effective in preventing *S. aureus* biofilms formation, however none of the nucleases were effective in disrupting mature biofilms (Kaplan et al., 2012).

3.2.6. Phytochemicals

Plant extracts and plant derived compounds have been used from the ancient times to combat infectious diseases in humans in different parts of the world. Plant derived compounds, including several polyphenolic compounds and flavonoids, exhibit wide range of biological activities such as anti-inflammatory, anti-allergic, hepatoprotective, anti-thrombotic, anti-carcinogenic and vasodilatory actions. Many of these plant-derived compounds also have weak antibiotic activity that is several orders of magnitudes weaker than that of common antibiotics produced by bacteria and fungi. Despite their weaker antibiotic activity some of these phytochemicals have potent anti-biofilm activity.

Extracts of the plants *Kaempferia rotunda* L., *Caesalpinia sappan* L., *Cinnamomum burmanii* Nees ex Bl., *C. sintoc* L., and *Nymphaea nouchali*, *L. Chilense, Tagetes minuta, T. absinthioides,* and *L. Divaricata* have been found to possess anti-biofilm activities (Pratiwi et al., 2015; Romero et al., 2016). Apart from these extracts several specific phytochemicals such as Ferulic acid [Source plant: *Ferula foetida* (Asafoetida)], Tannic

Table 1
In vivo validated *S. aureus* biofilm treatment strategies.

Compounds	Animal	Experimental outcome	Reference
Rifampicin	BALB/c mice	*S. aureus* Xen 29 pre-colonized catheter segments were implanted subcutaneously. Seven days post infection, rifampicin was administered intraperitoneally every 12 h for 4 consecutive days. 90% reduction in *S. aureus* biofilm was observed.	(Kadurugamuwa et al., 2003)
Daptomycin	Sprague-Dawley(SD) rats	Catheters were inserted in the jugular vein and were inoculated with *S. aureus*. Treatment with 5 mg/ml daptomycin as catheter lock solution along with systemic daptomycin (s.c.) for four consecutive days led to complete clearance of established biofilms.	(Van Praagh et al., 2011)
ML:8 (Major constituent: caprylic acid) and Citrox	SD rats	The preimplanted jugular vein catheters were inoculated with *S. aureus* strain USA300. Five days post infection, antibiotic lock therapy (ALT) with 50 µl ML:8 or Citrox was instilled into the lumen of the catheter daily for 4 days. Rats also received a subcutaneous injections of vancomycin (50 mg/kg of body weight) every 12 h throughout the duration of the experiment to prevent systemic infection. Both ML:8 and citrox were prevented *S. aureus* biofilm formation.	(Hogan et al., 2016)
Selenocyanatodiacetic acid (SCAA)	BALB/c mice.	SCAA coated hemodialysis catheters (HDC) were inserted subcutaneously on the back of the mouse and *S. aureus* AH133 was injected in the area of inserted catheter. *S. aureus*biofilm formation on SCAA coated HDC catheter was inhibited.	(Tran et al., 2012)
4-Benzylamino) cyclohexyl 2-hydroxycinnamate (BCHA)	Wistar rats	*S. aureus* precolonized polyethylene catheter was inserted into the urinary bladder of day 7 pregnant rat. Treatment with gentamicin and BCHA resulted in marked inhibition of biofilm development.	(Balamurugan et al., 2015)
Gentamicin-EDTA	CD/SD rats	*S. aureus* biofilms on totally implantable venous access ports (TIVAP) were treated with Gentamicin-EDTA ALT for 5 days in conjunction with systemic vancomycin injections. Treatment with gentamicin- EDTA ALT prevented initial adherence of *S. aureus* with TIVAP.	(Chauhan et al., 2012)
Nitric oxide-releasing nanoparticles (NO Np)	SD rats	Heparinized polyethylene catheters were inserted above the right atrium and were instilled with *S. aureus* strain USA300. 24 h postinfection 5 mg/ml of NO Np was injected to the catheter lumen. NO Np treated catheters exhibited low bacterial burden compared to untreated catheter.	(Mihu et al., 2017)
Surface modification of commercial (TIVAP) using methylcellulose (MeCe) and polyethylene glycol (PEG) derivatives	CD/SD rats	TIVAP surface modified with MeCe and PEG derivatives were implanted in rats and were infected with *S. aureus*. Modification of the TIVAP by MeCe and PEG derivatives inhibited initial surface adhesion of *S. aureus*.	(Chauhan et al., 2014)
Lysostaphin	CF-1 mice	Catheters were placed in the jugular vein. i) A single pre-instillation of lysostaphin (10 mg/kg) in catheters completely protected catheterized mice from a subsequent biofilm infection. ii) Lysostaphin (15 mg/kg) and nafcillin (50 mg/kg) three times a day for 4 days eradicated established *S. aureus* biofilms from implanted catheters.	(Kokai-Kun et al., 2009)
Mixture of trimethoprim + ethanol + and calcium Ethylenediaminetetraacetic acid	New Zealand white rabbits	Silicone catheter placed in the right external jugular vein, was inoculated with *S. aureus* and allowed to form intralumenal biofilm. ALT for 2 h/day for 7 days led to significant reduction of bacterial burden within the catheters.	(Chandra et al., 2018)
Silver nanoparticles (Ag Np)	SD rats	Medullary cavity of rat hind leg was inoculated with *S. aureus* ATCC 35984 and Ag Np immobilized or pure Titanium Kirschner wire was inserted into the cavity. Ag-Np was found to reduce the risk of implant associated peri prosthetic infection (PPI).	(Qin et al., 2014)
Shock waves	BALB/c mice	*S. aureus* was inoculated at the suture site generated by small incision made in the abdomen. Three days post infection, the mice were treated with ciprofloxacin alone, shock wave treatment alone, or shock wave (once per day) and ciprofloxacin for next 3 days. Shock waves treated mice biofilms became more susceptible to ciprofloxacin, while ciprofloxacin alone was ineffective in treating the suture site infection.	(Gnanadhas et al., 2015)
RNA III inhibiting peptide (RIP)	Wistar rats	Dacron grafts were soaked in solutions containing RIP and different antibiotics (mupirocin, quinupristin-dalfopristin, levofloxacin and rifampin) for 20 minutes prior to implantation in the subcutaneous pocket of Wistar rats. *S. aureus* strains were inoculated on the grafts. Combination of antibiotic and RIP displayed significant reduction in the *S. aureus* biofilm formation, compared to RIP and the antibiotics alone.	(Giacometti et al., 2003)
Antimicrobial peptide 16P-AC	SD rats	0.8 cm sterile stents were inserted into the rat bladders and were infected with *S. aureus*. Rats were treated using 16P-AC. The biofilm formation in the stents was significantly decreased.	(Zhou et al., 2016)

(*continued on next page*)

Table 1 (*continued*)

Compounds	Animal	Experimental outcome	Reference
Antimicrobial peptide Bac8c^{2,5Leu}	SD rats	Pre-implanted Rat Jugular Vein Catheters were infected with *S. aureus* strain USA300lux. Administration of D-Bac8c^{2,5Leu}as catheter lock solution along with systemic vancomycin injection for 5 days was highly effective in eradicating *S. aureus* biofilm infection.	(Zapotoczna et al., 2017)

acid [Source plants: Quercus species (Oak), *Camellia sinensis* (Tea)] (Payne et al., 2013), Ellagic acid [Source plants: Rubus idaeus (Raspberry), Fragaria ananassa (Strawberry), Rubus *fruticosus* (Blackberries), *Punicagranatum* (Pomegranate)] (Fontaine et al., 2017; Quave et al., 2012), gallic acid [Source plants: Quercus species (Oak)] (Liu et al., 2017; Luís et al., 2014), caffeic acid [Source plants: *Coffea* species] (Luís et al., 2014), chlorogenic acid [Source plants: *Coffea* species and *Camellia sinensis*] (Luís et al., 2014), Xanthohumol [Source plant: *Humulus lupulus*] (Rozalski et al., 2013), genistein [Source plant: *Genistatinctoria*] (Morán et al., 2014), protocatechuic acid [Source plant: *Camellia sinensis*] (Morán et al., 2014), p-hydroxybenzoic acid [Source plants: *Vitexagnus-castus* and *Hypericum perforatum*] (Morán et al., 2014), plumbagin [Source plants: *Plumbago zeylanica*] (Nair et al., 2016), Protocatechuic acid [Source plant: *Hibiscus sabdariffa*], Genistein [Source plants: *Flemingia vestita* and *F. macrophylla*], resveratrol [Source plants: Rubus idaeus (raspberry), *Vitis vinifera* (Grapes)], Sanguinarine [Source plant: *Sanguinaria canadensis* (bloodroot)] (Morán et al., 2014), Baicalein [Source plant: *Scutellaria baicalensis*] (Chen et al., 2016) and Hamamelitannin [Source plant: *Hamamelis virginiana* (witch hazel)] (Brackman et al., 2016) have been studied for their biofilm inhibitory properties. Most of the plant derivatives hold great promise to tackle *S. aureus* biofilm infections; however they require further *in vivo* experimental validations as all the data obtained from plant derived compounds are based on *in vitro* results. Few recent studies considered the combinatorial use of antibiotics and phytochemicals against *S. aureus* biofilms. Synergism was observed between combinations of oxacillin with xanthohumol (Rozalski et al., 2013); ciprofloxacin with quinine, pyrrolidine or reserpine; erythromycin with pyrrolidine; tetracycline with pyrrolidineormorin (Abreu et al., 2016) and vancomycin (VAN) with baicalein or hamamelitannin (Brackman et al., 2016; Chen et al., 2016).

3.2.7. Staphylococcal phages

Another potential therapeutic approach is phage therapy using staphylococcal phages. Phages are naturally occurring viruses that infect bacteria. Though quite popular in the Eastern European countries, phage therapy never gained enough importance in the rest of the world. However, they offer certain benefits over antibiotics. Bacteriophages can effectively target specific bacteria without affecting the commensals and even can kill the antibiotic resistant bacteria (Lin et al., 2017; Mattila et al., 2015). Even though bacteriophages are larger than antibiotics, they are reported to infect biofilm embedded bacteria and therefore are more effective than antibiotics in killing bacteria within the biofilms (Abedon, 2015; Harper et al., 2014).

Lytic phages with an ability to replicate inside the host are best suited for therapeutic use. Polyvalent phage K was acclaimed to inhibit biofilm formation of *S. aureus* (Lungren et al., 2013). Phage cocktails were also used to target staphylococcal biofilms. Alves et al., demonstrated that a combination of BacteriophageK and DRA88 (a broad host range phage) can effectively reduce *S. aureus* biofilm biomass within 48 h (Alves et al., 2014). Further, Drilling et al., used a phage cocktail of unknown composition which reduced biofilm of *S. aureus* (Drilling et al., 2014).

3.2.8. Antibodies

Numerous efforts were made to develop vaccines and therapeutic antibodies that can prevent and treat *S. aureus* infections. Attempts were made to use capsular polysaccharide (type 5 and 8), clumping factor-A and B, fibronectin binding protein, ABC transporter and amidase *etc* as vaccine candidates; and clumping factor A, adenosine triphosphate binding cassette transporter and teichoic acids *etc* as therapeutic antibodies (Nair et al., 2015). The efficacy of these antigens and antibodies was extensively tested in *S. aureus* induced sepsis model animals. However, little work has been carried out to design vaccines or therapeutic antibodies against *S. aureus* biofilm infections. In one study Lam et al, tested affinity-purified polyclonal antibodies, for their biofilm inhibitory properties against PhnD antigen (phosphonate ABC transporter substrate binding protein) and have found that the PhnD-specific antibodies blocked *S. aureus* biofilm development at the initial attachment and aggregation stages. PhnD-specific antibodies were also found to serve as opsonins that enhance biofilm engulfment by neutrophils (Lam et al., 2014). In other studies Xiong and Estellésa et al., used a human monoclonal antibody, TRL1068, against *S. aureus* biofilm. TRL1068 has high affinity against DNABII proteins which participates in holding eDNA within the biofilm extracellular matrix. TRL1068 was able to disrupt preformed *S. aureus* biofilm under *in vitro* and *in vivo* conditions (Estellés et al., 2016; Xiong et al., 2017). Additionally, synergism was observed between TRL1068 and the antibiotic daptomycin (Estellés et al., 2016).

3.3. Surface modifications of medical implants and catheters

Increased threat of *S. aureus* biofilm associated infections widened the search for alternative methods for preventing biofilm formation on the surface of implants and prosthetic devices. As biofilm formation is influenced by the physical properties of the biomaterials, cell surface dynamics is an important determining factor in the attachment of staphylococcal to biomaterial surfaces. Surface dynamics include the hydrophobicity, topology and electrostatic interactions. Hydrophobicity of the implant surface increases bacterial adhesion, especially on the plastic and metal surfaces. Surface modifications with methylcellulose, poly vinyl pyrrolidone, polyethylene glycol, poly ethylene oxide (PEO) and poly propylene oxide (PPO) were shown to have reduced staphylococcal adherence (Bridgett et al., 1992; Chauhan et al., 2014).

Commercially used standard biomaterials for implants include titanium, titanium alloy stainless steel, cobalt-chromium-molybdenum alloy and ultra-high-molecular-weight polyethylene. When the influence of surface morphology of biomaterials on *S. aureus* adhesion and biofilm formation was assessed, it was found that increased surface smoothness enhanced *S. aureus* attachment while increased surface roughness of the implant materials reduced *S. aureus* attachment (Lorenzetti et al., 2015; Singh et al., 2011). Creation of rough implant surfaces through nanopatterning using titanium oxide significantly reduced binding of *S. aureus* with the implant (Getzlaf et al., 2016).

Surface conjugation of biomaterials with antibiotics and other synthetic compounds are also under investigation. Antoci Jr et al demonstrated that titanium surfaces covalently derivatized with vancomycin resist *S. aureus* surface colonization (Antoci et al., 2007). Baveja et al reported that furanone coating inhibited staphylococcal adhesion and slime production on the surface of biomaterials (Baveja et al., 2004). Nano textured titania embedded with silver nanoparticles possess anti-biofilm activity (Mohandas et al., 2017; Qin et al., 2014).

3.4. Laser Shock waves (LSW), ultra sound (US) and photodynamic therapy (PDT)

With staphylococcal biofilms associated surgical site and prosthetic joints infections on rise, unconventional methods were also analysed. These include treatment with laser shock waves, ultra sound and photodynamic therapy which act by disrupting biofilms (Barra et al., 2015; Donnelly et al., 2009; Fu et al., 2013; García et al., 2015; Gnanadhas et al., 2015; Mai et al., 2016; Pérez-Laguna et al., 2017; Sharma et al., 2008). Laser shock waves are high energy waves travelling at supersonic speed and ultra sound is an oscillating sound which is beyond the upper limit of the human hearing range. Both these techniques were highly effective in disassembling biofilms and also augment antibiotic therapy. The *in vitro* and *in vivo* disruption of *S. aureus* biofilms by minimally invasive laser shock waves increase their susceptibility to the antibiotics ciprofloxacin and gentamicin (Gnanadhas et al., 2015; Qi et al., 2016). Similarly low frequency ultra sound treatment also enhanced the effect of antibiotic therapy on staphylococcal biofilms. Photodynamic therapy uses light of a particular wavelength to activate photosensitizing agents which produce reactive oxygen species that are deleterious to the bacteria. Photosensitizers such as malachite green (Rosa et al., 2015), methylene blue (Rosa et al., 2015), sinoporphyrin sodium (Mai et al., 2016), toluidine blue O (Rosa et al., 2015; Sharma et al., 2008), chlorin e6 (Park et al., 2012) and 5-aminolevulinic acid (Zhang et al., 2017) were effective in staphylococcal biofilm disruption. The anti-biofilm effect of 5-aminolevulinic acid was found to increase when used in combination with netilmicin, vancomycin, and cefaclor antibiotics (Zhang et al., 2017).

4. Conclusions and perspective

S. aureus is included in the 'high' priority category of World health organization list of antibiotic resistant pathogens that pose a threat to human health. One of the strongest survival tactics used by *S. aureus* is the biofilm formation which is difficult to treat using antibiotics. The standard treatment of *S. aureus* biofilm infections include removal of the infected foreign bodies and systemic or topical antibiotic administration in high dosage, which requires hospital readmission, additional surgical procedures leading to tremendous increase in treatment costs. Therefore, research on the prevention and treatment of the staphylococcal biofilm claims utmost importance. Recent research reports highlight several innovative preventive and therapeutic strategies that could be adopted to tackle *S. aureus* biofilm infections, which includes use of appropriate antibiotic combinations, phytochemicals, enzymes, AMPs, sulfahydrl compounds, nanoparticles, phages, and metal chelators. Unfortunately, only a few of these anti-biofilm agents were tested under *in vivo* conditions (Table 1). The vast majority of the *in vitro* identified anti-biofilm agents require further *in vivo* verification and validation, as many of these *in vitro* identified anti-biofilm agents do have certain disadvantages. For example, even though phage therapy is advantageous for the reason that phages are in general bacteria specific but their biological origin hinders their therapeutic use. Metal chelators and sulfhydryl agents showed excellent anti-biofilm activities, but are unsuitable for systemic application. For example, animal studies demonstrated the chelating agent EDTA is cytotoxic and weakly genotoxic. Sulfadryl agent dithiothreitol may cause skin irritation and organ toxicity. Metallic nanoparticles at higher dose may exert toxicity as was observed for silver nanoparticles. *In vivo* rat model experiments demonstrated that high doses of silver nanoparticles exert both cytotoxic and genotoxic effects (Wen et al., 2017). Nucleases and enzymes can disrupt the biofilm extracellular matrix, but they are protein in nature and their systemic application is again restricted due to their possibility of activating immune responses. Hence, the above mentioned anti-biofilm agents may only be considered to be developed as antimicrobial catheter lock solutions. Antimicrobial catheter lock (ACL) solutions are used to fill the catheter tubes when they are not in use. A number of antibiotics and other compounds (Daptomycin, Gentamicin-EDTA, ML:8-Citrox and trimethoprim-ethanol-calcium-EDTA) have been tested recently as ACL solutions in animal models (Chauhan et al., 2012; Hogan et al., 2016; Van Praagh et al., 2011). Other than the above-mentioned anti-biofilm agents, few synthetic compounds have also been reported to inhibit *S. aureus* biofilm formation. 11-triphenylsilyl-10,11-dihydrocinchonidine (Skogman et al., 2012), 1,3- Bis-(2-hydroxy-phenyl)-propenone (Bozic et al., 2014), 4-(Benzylamino)cyclohexyl 2-hydroxycinnamate, 2-[(Methylamino)methyl]phenol (Balamurugan et al., 2015), Ethyl gallate, Hexyl gallate, Octadecyl gallate, (*E*)-*N*′-(4-bromo-benzylidene)benzohydrazide, (2*E*)-1-(3′,4′-dimethoxyphenyl)-3-(2-naphthyl)-2-propen-1-one down-regulate biofilm development of *S. aureus* (de Lima Pimenta et al., 2013). Further research is necessary for the development of most appropriate anti-biofilm agents for systemic applications. In this regard cyto-compatible and biocompatible phytochemicals, drug loaded polymeric nanoparticles and repurposed drugs may act as better therapeutics. A number of phytochemicals also acts in synergy with antibiotics. Additionally, laser shock waves, ultra sound and photodynamic therapy act by disrupting biofilms and demonstrated synergism with antibiotics. Under the present conditions, development of an effective vaccine and therapeutic antibodies against *S. aureus* biofilms are highly desirable. While developing biofilm dispersive compounds it is necessary to recall that catheters and other biomaterials get coated with host proteins soon after implantation. Specific *S. aureus* proteins such as, Atl, FnBPA and FnBPB, ClfA, ClfB and teichoic acids play vital role in initial attachment of *S. aureus* with host protein coated surfaces. Thus, prevention of initial attachment of *S. aureus* with catheters and other biomaterials could be another therapeutic strategy. To summarize, in this review we have discussed the recent developments in the *S. aureus* biofilm prevention and treatment options, and their possible use and limitations. However, most findings described in this review require further *in vivo* validation. So far, only one clinical trial was registered where the efficacy of *S. epidermidis* serine protease (Esp) protein was intended to test for its *S. aureus* biofilm disrupting activities in venous ulcer patients wounds, but this study was prematurely withdrawn due to the unavailability of sufficient amount of purified Esp protein (NCT01646502). Further studies are necessary to identify more suitable biofilm disrupting agents.

Conflict of interest

No conflicts.

Acknowledgements

This work was supported by the research grant from Indian Council Of Medical Research, India to LB (ICMR AMR/14/2011-ECD1) and senior research fellowship from Council of Scientific & Industrial Research (CSIR), India, to MKS (09/963 (0037) 2Kl7- EMR-I). We thank the Centre for Nanoscience and Molecular Medicine, Kochi, Amrita VishwaVidyapeetham, India for infrastructural support.

References

Abedon, S.T., 2015. Ecology of anti-biofilm agents II: bacteriophage exploitation and biocontrol of biofilm bacteria. Pharmaceuticals 8, 559–589. https://doi.org/10.3390/ph8030559.

Abraham, N.M., Lamlertthon, S., Fowler, V.G., Jefferson, K.K., 2012. Chelating agents exert distinct effects on biofilm formation in *Staphylococcus aureus* depending on strain background: role for clumping factor B. J. Med. Microbiol. 61, 1062–1070. https://doi.org/10.1099/jmm.0.040758-0.

Abreu, A.C., Saavedra, M.J., Simões, L.C., Simões, M., 2016. Combinatorial approaches with selected phytochemicals to increase antibiotic efficacy against *Staphylococcus aureus* biofilms. Biofouling 32, 1103–1114. https://doi.org/10.1080/08927014.2016.1232402.

Aguinagalde, L., Díez-Martínez, R., Yuste, J., Royo, I., Gil, C., Lasa, Í., Martín-Fontecha, M., Marín-Ramos, N.I., Ardanuy, C., Liñares, J., García, P., García, E., Sánchez-Puelles, J.M., 2015. Auranofin efficacy against MDR *Streptococcus pneumoniae* and

Staphylococcus aureus infections. J. Antimicrob. Chemother. 70, 2608–2617. https://doi.org/10.1093/jac/dkv163.
Alves, D.R., Gaudion, A., Bean, J.E., Perez Esteban, P., Arnot, T.C., Harper, D.R., Kot, W., Hansen, L.H., Enright, M.C., Jenkins, A.T.A., 2014. Combined use of bacteriophage K and a novel bacteriophage to reduce *Staphylococcus aureus* biofilm formation. Appl. Environ. Microbiol. 80, 6694–6703. https://doi.org/10.1128/AEM.01789-14.
Antoci, V., King, S.B., Jose, B., Parvizi, J., Zeiger, A.R., Wickstrom, E., Freeman, T.A., Composto, R.J., Ducheyne, P., Shapiro, I.M., Hickok, N.J., Adams, C.S., 2007. Vancomycin covalently bonded to titanium alloy prevents bacterial colonization. J. Orthop. Res. 25, 858–866. https://doi.org/10.1002/jor.20348.
Applerot, G., Lellouche, J., Perkas, N., Nitzan, Y., Gedanken, A., Banin, E., 2012. ZnO nanoparticle-coated surfaces inhibit bacterial biofilm formation and increase antibiotic susceptibility. RSC Adv. 2, 2314. https://doi.org/10.1039/c2ra00602b.
Archer, N.K., Mazaitis, M.J., Costerton, J.W., Leid, J.G., Powers, M.E., Shirtliff, M.E., 2011a. *Staphylococcus aureus* biofilms. Virulence 2, 445–459. https://doi.org/10.4161/viru.2.5.17724.
Archer, N.K., Mazaitis, M.J., William Costerton, J., Leid, J.G., Powers, M.E., Shirtliff, M.E., 2011b. *Staphylococcus aureus* biofilms: properties, regulation and roles in human disease. Virulence. https://doi.org/10.4161/viru.2.5.17724.
Arciola, C.R., Campoccia, D., Ravaioli, S., Montanaro, L., 2015. Polysaccharide intercellular adhesin in biofilm: structural and regulatory aspects. Front. Cell. Infect. Microbiol. 5. https://doi.org/10.3389/fcimb.2015.00007.
Balamurugan, P., Hema, M., Kaur, G., Sridharan, V., Prabu, P.C., Sumana, M.N., Princy, S.A., 2015. Development of a biofilm inhibitor molecule against multidrug resistant *Staphylococcus aureus* associated with gestational urinary tract infections. Front. Microbiol. 6, 832. https://doi.org/10.3389/fmicb.2015.00832.
Barbu, E.M., Mackenzie, C., Foster, T.J., Höök, M., 2014. SdrC induces staphylococcal biofilm formation through a homophilic interaction. Mol. Microbiol. 94, 172–185. https://doi.org/10.1111/mmi.12750.
Barra, F., Roscetto, E., Soriano, A.A., Vollaro, A., Postiglione, I., Pierantoni, M.G., Palumbo, G., Catania, M.R., 2015. Photodynamic and antibiotic therapy in combination to fight biofilms and resistant surface bacterial infections. Int. J. Mol. Sci. 16, 20417–20430. https://doi.org/10.3390/ijms160920417.
Baveja, J.K., Willcox, M.D.P., Hume, E.B.H., Kumar, N., Odell, R., Poole-Warren, L.A., 2004. Furanones as potential anti-bacterial coatings on biomaterials. Biomaterials 25, 5003–5012. https://doi.org/10.1016/j.biomaterials.2004.02.051.
Boles, B.R., Horswill, A.R., 2008. *Agr*-mediated dispersal of *Staphylococcus aureus* biofilms. PLoS Pathog. 4, e1000052. https://doi.org/10.1371/journal.ppat.1000052.
Bose, J.L., Lehman, M.K., Fey, P.D., Bayles, K.W., 2012. Contribution of the *Staphylococcus aureus* Atl AM and GL murein hydrolase activities in cell division, autolysis, and biofilm formation. PLoS One 7, e42244. https://doi.org/10.1371/journal.pone.0042244.
Bozic, D.D., Milenkovic, M., Ivkovic, B., Cirkovic, I., 2014. Newly-synthesizedchalcones-inhibition of adherence and biofilm formation of methicillin-resistant *Staphylococcus aureus*. Braz. J. Microbiol. 45, 263–270.
Brackman, G., Breyne, K., De Rycke, R., Vermote, A., Van Nieuwerburgh, F., Meyer, E., Van Calenbergh, S., Coenye, T., 2016. The quorum sensing inhibitor Hamamelitannin increases antibiotic susceptibility of *Staphylococcus aureus* biofilms by affecting peptidoglycan biosynthesis and eDNA release. Sci. Rep. 6, 20321. https://doi.org/10.1038/srep20321.
Bradford, C., 2011. The use of commercially available α-Amylase compounds to inhibit and remove *Staphylococcus aureus* biofilms. Open Microbiol. J. 5, 21–31. https://doi.org/10.2174/1874285801105010021.
Breger, J., Fuchs, B.B., Aperis, G., Moy, T., Ausubel, F.M., Mylonakis, E., 2007. Antifungal chemical compounds identified using a *C. elegans* pathogenicity assay. PLoS Pathog. 3, e18. https://doi.org/10.1371/journal.ppat.0030018.
Bridgett, M.J., Davies, M.C., Denyer, S.P., 1992. Control of staphylococcal adhesion to polystyrene surfaces by polymer surface modification with surfactants. Biomaterials 13, 411–416. https://doi.org/10.1016/0142-9612(92)90159-L.
Britto, C.J., Cohn, L., 2015. Bactericidal/Permeability-increasing protein fold-containing family member A1 in airway host protection and respiratory disease. Am. J. Respir. Cell Mol. Biol. 52, 525–534. https://doi.org/10.1165/rcmb.2014-0297RT.
Caroli, A., Simeoni, S., Lepore, R., Tramontano, A., Via, A., 2012. Investigation of a potential mechanism for the inhibition of SmTGR by Auranofin and its implications for Plasmodium falciparum inhibition. Biochem. Biophys. Res. Commun. 417, 576–581. https://doi.org/10.1016/j.bbrc.2011.12.009.
Chandra, J., Long, L., Isham, N., Mukherjee, P.K., DiSciullo, G., Appelt, K., Ghannoum, M.A., 2018. *In Vitro* and *In Vivo* Activity of a Novel Catheter Lock Solution against Bacterial and Fungal Biofilms. Antimicrob. Agents Chemother. 62https://doi.org/10.1128/AAC.00722-18. 8. pii: e00722–18.
Chaudhari, P.R., Masurkar, S.A., Shidore, V.B., Kamble, S.P., 2012. Effect of biosynthesized silver nanoparticles on *Staphylococcus aureus* biofilm quenching and prevention of biofilm formation. Int. J. Pharma Bio Sci. 3, 222–229. https://doi.org/10.1007/BF03353689.
Chauhan, A., Lebeaux, D., Ghigo, J.M., Beloin, C., 2012. Full and broad-spectrum in vivo eradication of catheter-associated biofilms using gentamicin-EDTA antibiotic lock therapy. Antimicrob. Agents Chemother. 56, 6310–6318. https://doi.org/10.1128/AAC.01606-12.
Chauhan, A., Bernardin, A., Mussard, W., Kriegel, I., Estève, M., Ghigo, J.M., Beloin, C., Semetey, V., 2014. Preventing biofilm formation and associated occlusion by biomimetic glycocalyxlike polymer in central venous catheters. J. Infect. Dis. 210, 1347–1356. https://doi.org/10.1093/infdis/jiu249.
Chen, C., Krishnan, V., Macon, K., Manne, K., Narayana, S.V.L., Schneewind, O., 2013. Secreted proteases control autolysin-mediated biofilm growth of Staphylococcus aureus. J. Biol. Chem. 288, 29440–29452. https://doi.org/10.1074/jbc.M113.502039.
Chen, Y., Liu, T., Wang, K., Hou, C., Cai, S., Huang, Y., Du, Z., Huang, H., Kong, J., Chen, Y., 2016. Baicalein inhibits *Staphylococcus aureus* biofilm formation and the quorum sensing system in vitro. PLoS One 11, e0153468. https://doi.org/10.1371/journal.pone.0153468.
Chessa, D., Ganau, G., Spiga, L., Bulla, A., Mazzarello, V., Campus, G.V., Rubino, S., 2016. *Staphylococcus aureus* and Staphylococcus epidermidis virulence strains as causative agents of persistent infections in breast implants. PLoS One 11, e0146668. https://doi.org/10.1371/journal.pone.0146668.
Chirullo, B., Sgarbanti, R., Limongi, D., Shytaj, I.L., Alvarez, D., Das, B., Boe, A., DaFonseca, S., Chomont, N., Liotta, L., Petricoin, E.I., Norelli, S., Pelosi, E., Garaci, E., Savarino, A., Palamara, A.T., 2013. A candidate anti-HIV reservoir compound, auranofin, exerts a selective' anti-memory' effect by exploiting the baseline oxidative status of lymphocytes. Cell Death Dis. 4https://doi.org/10.1038/cddis.2013.473. e944.
Chung, P.Y., Toh, Y.S., 2014. Anti-biofilm agents: recent breakthrough against multi-drug resistant Staphylococcus aureus. Pathog. Dis. 70, 231–239. https://doi.org/10.1111/2049-632X.12141.
Corrigan, R.M., Rigby, D., Handley, P., Foster, T.J., 2007. The role of *Staphylococcus aureus* surface protein SasG in adherence and biofilm formation. Microbiology 153, 2435–2446. https://doi.org/10.1099/mic.0.2007/006676-0.
Corsello, S.M., Bittker, J.A., Liu, Z., Gould, J., McCarren, P., Hirschman, J.E., Johnston, S.E., Vrcic, A., Wong, B., Khan, M., Asiedu, J., Narayan, R., Mader, C.C., Subramanian, A., Golub, T.R., 2017. The Drug Repurposing Hub: a next-generation drug library and information resource. Nat. Med. 23, 405–408. https://doi.org/10.1038/nm.4306.
Cramton, S.E., Gerke, C., Schnell, N.F., Nichols, W.W., Götz, F., 1999. The intercellular adhesion (*ica*) locus is present in *Staphylococcus aureus* and is required for biofilm formation. Infect. Immun. 67, 5427–5433.
Cucarella, C., Tormo, M.Á., Úbeda, C., Trotonda, M.P., Monzón, M., Peris, C., Amorena, B., Lasa, Í., Penadés, J.R., 2004. Role of biofilm-associated protein Bap in the pathogenesis of bovine *Staphylococcus aureus*. Infect. Immun. 72, 2177–2185. https://doi.org/10.1128/IAI.72.4.2177-2185.2004.
Dawgul, M., Baranska-Rybak, W., Piechowicz, L., Bauer, M., Neubauer, D., Nowicki, R., Kamysz, W., 2016. The antistaphylococcal activity of citropin 1.1 and temporin a against planktonic cells and biofilms formed by isolates from patients with atopic dermatitis: an assessment of their potential to induce microbial resistance compared to conventional antimicrobials. Pharmaceuticals (Basel) 9, e30. https://doi.org/10.3390/ph9020030.
De Leo, F., Diep, B., Otto, M., 2009. Host defense and pathogenesis in *Staphylococcus aureus* infections. Infect. Dis. Clin. North Am. 23, 17–34. https://doi.org/10.1016/j.idc.2008.10.003.Host.
de Lima Pimenta, A., Chiaradia-Delatorre, L.D., Mascarello, A., de Oliveira, K.A., Leal, P.C., Yunes, R.A., de Aguiar, C.B., Tasca, C.I., Nunes, R.J., Smânia, A.Jr., 2013. Synthetic organic compounds with potential for bacterial biofilm inhibition, a path for the identification of compounds interfering with quorum sensing. Int. J. Antimicrob. Agents 42, 519–523. https://doi.org/10.1016/j.ijantimicag.2013.07.006.
Dean, S.N., Bishop, B.M., van Hoek, M.L., 2011. Natural and synthetic cathelicidin peptides with anti-microbial and anti-biofilm activity against Staphylococcus aureus. BMC Microbiol. 11, 114. https://doi.org/10.1186/1471-2180-11-114.
Deftereos, S.N., Andronis, C., Friedla, E.J., Persidis, A., Persidis, A., 2011. Drug repurposing and adverse event prediction using high-throughput literature analysis. Wiley Interdiscip. Rev. Syst. Biol. Med. 3, 323–334. https://doi.org/10.1002/wsbm.147.
Diamond, G., Beckloff, N., Weinberg, A., Kisich, K.O., 2009. The roles of antimicrobial peptides in innate host defense. Curr. Pharm. Des. 15, 2377–2392.
Donlan, R.M., 2002. Biofilms: microbial life on surfaces. Emerg. Infect. Dis. 8, 881–890. https://doi.org/10.3201/eid0809.020063.
Donnelly, R.F., Cassidy, C.M., Loughlin, R.G., Brown, A., Tunney, M.M., Jenkins, M.G., McCarron, P.A., 2009. Delivery of Methylene Blue and meso-tetra (N-methyl-4-pyridyl) porphine tetra tosylate from cross-linked poly(vinyl alcohol) hydrogels: a potential means of photodynamic therapy of infected wounds. J. Photochem. Photobiol. B Biol. 96, 223–231. https://doi.org/10.1016/j.jphotobiol.2009.06.010.
Drilling, A., Morales, S., Jardeleza, C., Vreugde, S., Speck, P., Wormald, P.J., 2014. Bacteriophage reduces biofilm of *Staphylococcus aureus* ex vivo isolates from chronic rhinosinusitis patients. Am. J. Rhinol. Allergy 28, 3–11. https://doi.org/10.2500/ajra.2014.28.4001.
Elchinger, P.H., Delattre, C., Faure, S., Roy, O., Badel, S., Bernardi, T., Taillefumier, C., Michaud, P., 2014. Effect of proteases against biofilms of *Staphylococcus aureus* and staphylococcus epidermidis. Lett. Appl. Microbiol. 59, 507–513. https://doi.org/10.1111/lam.12305.
Estellés, A., Woischnig, A.K., Liu, K., Stephenson, R., Lomongsod, E., Nguyen, D., Zhang, J., Heidecker, M., Yang, Y., Simon, R.J., Tenorio, E., Ellsworth, S., Leighton, A., Ryser, S., Gremmelmaier, N.K., Kauvar, L.M., 2016. A high-affinity native human antibody disrupts biofilm from *Staphylococcus aureus* bacteria and potentiates antibiotic efficacy in a mouse implant infection model. Antimicrob. Agents Chemother. 60, 2292–2301. https://doi.org/10.1128/AAC.02588-15.
Fenton, M., Keary, R., McAuliffe, O., Ross, R.P., O'Mahony, J., Coffey, A., 2013. Bacteriophage-derived peptidase CHAPK eliminates and prevents staphylococcal biofilms. Int. J. Microbiol., 625341. https://doi.org/10.1155/2013/625341. 2013.
Fitzpatrick, F., Humphreys, H., O'Gara, J.P., 2005. Evidence for icaADBC -independent biofilm development mechanism in methicillin-resistant *Staphylococcus aureus* clinical isolates. J. Clin. Microbiol. 43, 1973–1976. https://doi.org/10.1128/JCM.43.4.1973.
Flemming, H.C., Wingender, J., Szewzyk, U., Steinberg, P., Rice, S.A., Kjelleberg, S., 2016. Biofilms: an emergent form of bacterial life. Nat. Rev. Microbiol. 14, 563–575.

https://doi.org/10.1038/nrmicro.2016.94.
Fluckiger, U., Ulrich, M., Steinhuber, A., Döring, G., Mack, D., Landmann, R., Goerke, C., Wolz, C., 2005. Biofilm formation, *ica*ADBC transcription, and polysaccharide intercellular adhesin synthesis by *staphylococci* in a device-related infection model. Infect. Immun. 73, 1811–1819. https://doi.org/10.1128/IAI.73.3.1811-1819.2005.
Fontaine, B.M., Nelson, K., Lyles, J.T., Jariwala, P.B., García-Rodriguez, J.M., Quave, C.L., Weinert, E.E., 2017. Identification of ellagic acid rhamnoside as a bioactive component of a complex botanical extract with anti-biofilm activity. Front. Microbiol. 8, 496. https://doi.org/10.3389/fmicb.2017.00496.
Fu, X.-J., Fang, Y., Yao, M., 2013. Antimicrobial photodynamic therapy for methicillin-resistant *Staphylococcus aureus* infection. Biomed Res. Int., 159157. https://doi.org/10.1155/2013/159157. 2013.
García, I., Ballesta, S., Gilaberte, Y., Rezusta, A., Pascual, Á., 2015. Antimicrobial photodynamic activity of hypericin against methicillin-susceptible and resistant *Staphylococcus aureus* biofilms. Future Microbiol. 10, 347–356. https://doi.org/10.2217/fmb.14.114.
Getzlaf, M.A., Lewallen, E.A., Kremers, H.M., Jones, D.L., Bonin, C.A., Dudakovic, A., Thaler, R., Cohen, R.C., Lewallen, D.G., van Wijnen, A.J., 2016. Multi-disciplinary antimicrobial strategies for improving orthopaedic implants to prevent prosthetic joint infections in hip and knee. J. Orthop. Res. 34, 177–186. https://doi.org/10.1002/jor.23068.
Giacometti, A., Cirioni, O., Gov, Y., Ghiselli, R., Del Prete, M.S., Mocchegiani, F., Saba, V., Orlando, F., Scalise, G., Balaban, N., Dell'Acqua, G., 2003. RNA III inhibiting peptide inhibits *in vivo* biofilm formation by drug-resistant *Staphylococcus aureus*. Antimicrob. Agents Chemother. 47, 1979–1983.
Gilan, I., Sivan, A., 2013. Effect of proteases on biofilm formation of the plastic-degrading actinomycete Rhodococcus ruber C208. FEMS Microbiol. Lett. 342, 18–23. https://doi.org/10.1111/1574-6968.12114.
Gnanadhas, D.P., Elango, M., Janardhanraj, S., Srinandan, C.S., Datey, A., Strugnell, R.A., Gopalan, J., Chakravortty, D., 2015. Successful treatment of biofilm infections using shock waves combined with antibiotic therapy. Sci. Rep. 5, 17440. https://doi.org/10.1038/srep17440.
Gordya, N., Yakovlev, A., Kruglikova, A., Tulin, D., Potolitsina, E., Suborova, T., Bordo, D., Rosano, C., Chernysh, S., 2017. Natural antimicrobial peptide complexes in the fighting of antibiotic resistant biofilms: *calliphora vicina* medicinal maggots. PLoS One 12, e0173559. https://doi.org/10.1371/journal.pone.0173559.
Günther, F., Wabnitz, G.H., Stroh, P., Prior, B., Obst, U., Samstag, Y., Wagner, C., Hänsch, G.M., 2009. Host defence against *Staphylococcus aureus* biofilms infection: phagocytosis of biofilms by polymorphonuclear neutrophils (PMN). Mol. Immunol. 46, 1805–1813. https://doi.org/10.1016/j.molimm.2009.01.020.
Gutiérrez, D., Ruas-Madiedo, P., Martínez, B., Rodríguez, A., García, P., 2014. Effective removal of staphylococcal biofilms by the endolysin LysH5. PLoS One 9, e107307. https://doi.org/10.1371/journal.pone.0107307.
Gwisai, T., Hollingsworth, N.R., Cowles, S., Tharmalingam, N., Mylonakis, E., Fuchs, B.B., Shukla, A., 2017. Repurposing niclosamide as a versatile antimicrobial surface coating against device-associated, hospital-acquired bacterial infections. Biomed. Mater. 12, 045010. https://doi.org/10.1088/1748-605X/aa7105.
Hall-Stoodley, L., Costerton, J., Stoodley, P., 2004. Bacterial biofilms: from the natural environment to infectious diseases. Nat. Rev. Microbiol. 2, 95–108. https://doi.org/10.1038/nrmicro821.
Harper, D., Parracho, H., Walker, J., Sharp, R., Hughes, G., Werthén, M., Lehman, S., Morales, S., 2014. Bacteriophages and biofilms. Antibiotics 3, 270–284. https://doi.org/10.3390/antibiotics3030270.
Hernandez, J.J., Pryszlak, M., Smith, L., Yanchus, C., Kurji, N., Shahani, V.M., Molinski, S.V., 2017. Giving drugs a second chance: overcoming regulatory and financial hurdles in repurposing approved drugs As Cancer therapeutics. Front. Oncol. 7, 273. https://doi.org/10.3389/fonc.2017.00273.
Hogan, S., Zapotoczna, M., Stevens, N.T., Humphreys, H., O'Gara, J.P., O'Neill, E., 2016. Eradication of *Staphylococcus aureus* catheter-related biofilm infections using ML:8 and Citrox. Antimicrob. Agents Chemother. 60, 5968–5975. https://doi.org/10.1128/AAC.00910-16.
Ibberson, C.B., Parlet, C.P., Kwiecinski, J., Crosby, H.A., Meyerholz, D.K., Horswill, A.R., 2016. Hyaluronan modulation impacts *Staphylococcus aureus* biofilm infection. Infect. Immun. 84, 1917–1929. https://doi.org/10.1128/IAI.01418-15.
Joo, H.S., Otto, M., 2012. Molecular basis of *in vivo* biofilm formation by bacterial pathogens. Chem. Biol. 19, 1503–1513. https://doi.org/10.1016/j.chembiol.2012.10.022.
Jorge, P., Grzywacz, D., Kamysz, W., Lourenço, A., Pereira, M.O., 2017. Searching for new strategies against biofilm infections: Colistin-AMP combinations against *Pseudomonas aeruginosa* and *Staphylococcus aureus* single- and double-species biofilms. PLoS One 12https://doi.org/10.1371/journal.pone.0174654. e0174654.
Kadurugamuwa, J.L., Sin, L.V., Yu, J., Francis, K.P., Kimura, R., Purchio, T., Contag, P.R., 2003. Rapid direct method for monitoring antibiotics in a mouse model of bacterial biofilm infection. Antimicrob. Agents Chemother. 47, 3130–3137. https://doi.org/10.1128/AAC.47.10.3130.
Kaplan, J.B., Velliyagounder, K., Ragunath, C., Rohde, H., Mack, D., Knobloch, J.K.M., Ramasubbu, N., 2004. Genes involved in the synthesis and degradation of matrix polysaccharide in Actinobacillus actinomycetemcomitans and Actinobacillus pleuropneumoniae biofilms. J. Bacteriol. 186, 8213–8220. https://doi.org/10.1128/JB.186.24.8213-8220.2004.
Kaplan, J.B., Lovetri, K., Cardona, S.T., Madhyastha, S., Sadovskaya, I., Jabbouri, S., Izano, E.A., 2012. Recombinant human DNase i decreases biofilm and increases antimicrobial susceptibility in staphylococci. J. Antibiot. (Tokyo) 65, 73–77. https://doi.org/10.1038/ja.2011.113.
Kiedrowski, M.R., Kavanaugh, J.S., Malone, C.L., Mootz, J.M., Voyich, J.M., Smeltzer, M.S., Bayles, K.W., Horswill, A.R., 2011. Nuclease modulates biofilm formation in community-associated methicillin-resistant Staphylococcus aureus. PLoS One 6, e26714. https://doi.org/10.1371/journal.pone.0026714.
Kokai-Kun, J.F., Chanturiya, T., Mond, J.J., 2009. Lysostaphin eradicates established *Staphylococcus aureus* biofilms in jugular vein catheterized mice. J. Antimicrob. Chemother. 64, 94–100. https://doi.org/10.1093/jac/dkp145.
Kolar, S.L., Antonio Ibarra, J., Rivera, F.E., Mootz, J.M., Davenport, J.E., Stevens, S.M., Horswill, A.R., Shaw, L.N., 2013. Extracellular proteases are key mediators of *Staphylococcus aureus* virulence via the global modulation of virulence-determinant stability. Microbiologyopen 2, 18–34. https://doi.org/10.1002/mbo3.55.
Kotulová, D., Slobodníková, L., 2010. Susceptibility of *Staphylococcus aureus* biofilms to vancomycin, gemtamicin and rifampin. Epidemiol. Mikrobiol. Imunol. 59, 80–87.
Lam, H., Kesselly, A., Stegalkina, S., Kleanthous, H., Yethon, J.A., 2014. Antibodies to PhnD inhibit staphylococcal biofilms. Infect. Immun. 82, 3764–3774. https://doi.org/10.1128/IAI.02168-14.
LaPlante, K.L., Mermel, L.A., 2009. In vitro activities of telavancin and vancomycin against biofilm-producing *Staphylococcus aureus*, *S. epidermidis*, and *Enterococcus faecalis* strains. Antimicrob. Agents Chemother. 53, 3166–3169. https://doi.org/10.1128/AAC.01642-08.
Laverty, G., Gorman, S.P., Gilmore, B.F., 2013. Biomolecular mechanisms of staphylococcal biofilm formation. Future Microbiol. 8, 509–524. https://doi.org/10.2217/fmb.13.7.
Le, K.Y., Dastgheyb, S., Ho, T.V., Otto, M., 2014. Molecular determinants of staphylococcal biofilm dispersal and structuring. Front. Cell. Infect. Microbiol. 4, 167. https://doi.org/10.3389/fcimb.2014.00167.
Lebeaux, D., Ghigo, J.-M., Beloin, C., 2014. Biofilm-related infections: bridging the gap between clinical management and fundamental aspects of recalcitrance toward Antibiotics. Microbiol. Mol. Biol. Rev. 78, 510–543. https://doi.org/10.1128/MMBR.00013-14.
Lellouche, J., Kahana, E., Elias, S., Gedanken, A., Banin, E., 2009. Antibiofilm activity of nanosized magnesium fluoride. Biomaterials 30, 5969–5978. https://doi.org/10.1016/j.biomaterials.2009.07.037.
Lellouche, J., Friedman, A., Gedanken, A., Banin, E., 2012a. Antibacterial and antibiofilm properties of yttrium fluoride nanoparticles. Int. J. Nanomedicine 7, 5611–5624. https://doi.org/10.2147/IJN.S37075.
Lellouche, J., Friedman, A., Lahmi, R., Gedanken, A., Banin, E., 2012b. Antibiofilm surface functionalization of catheters by magnesium fluoride nanoparticles. Int. J. Nanomedicine 7, 1175–1188. https://doi.org/10.2147/IJN.S26770.
Lewis, K., 2008. Multidrug tolerance of biofilms and persister cells. Curr. Top. Microbiol. Immunol. 322, 107–131. https://doi.org/10.1007/978-3-540-75418-3_6.
Li, J., Koh, J.J., Liu, S., Lakshminarayanan, R., Verma, C.S., Beuerman, R.W., 2017. Membrane active antimicrobial peptides: translating mechanistic insights to design. Front. Neurosci. 11 73. https://doi.org/10.3389/fnins.2017.00073.
Lin, D.M., Koskella, B., Lin, H.C., 2017. Phage therapy: an alternative to antibiotics in the age of multi-drug resistance. World J. Gastrointest. Pharmacol. Ther. 8, 162. https://doi.org/10.4292/wjgpt.v8.i3.162.
Lister, J.L., Horswill, A.R., 2014. *Staphylococcus aureus* biofilms: recent developments in biofilm dispersal. Front. Cell. Infect. Microbiol. 4, 178. https://doi.org/10.3389/fcimb.2014.00178.
Liu, M., Wu, X., Li, J., Liu, L., Zhang, R., Shao, D., Du, X., 2017. The specific anti-biofilm effect of gallic acid on *Staphylococcus aureus* by regulating the expression of the ica operon. Food Control 73, 613–618. https://doi.org/10.1016/j.foodcont.2016.09.015.
Lorenzetti, M., Dogša, I., Stošicki, T., Stopar, D., Kalin, M., Kobe, S., Novak, S., 2015. The influence of surface modification on bacterial adhesion to titanium-based substrates. ACS Appl. Mater. Interfaces 7, 1644–1651. https://doi.org/10.1021/am507148n.
Loughran, A.J., Atwood, D.N., Anthony, A.C., Harik, N.S., Spencer, H.J., Beenken, K.E., Smeltzer, M.S., 2014. Impact of individual extracellular proteases on *Staphylococcus aureus* biofilm formation in diverse clinical isolates and their isogenic sarA mutants. Microbiologyopen 3, 897–909. https://doi.org/10.1002/mbo3.214.
Luís, Â., Silva, F., Sousa, S., Duarte, A.P., Domingues, F., 2014. Antistaphylococcal and biofilm inhibitory activities of gallic, caffeic, and chlorogenic acids. Biofouling 30, 69–79. https://doi.org/10.1080/08927014.2013.845878.
Lundberg, F., Tegenfeldt, J.O., Montelius, L., Ransjö, U., Appelgren, P., Siesjö, P., Ljungh, Å., 1997. Protein depositions on one hydrocephalus shunt and on fifteen temporary ventricular catheters. Acta Neurochir. (Wien) 139 (8), 734–742. https://doi.org/10.1007/BF01420046.
Lungren, M.P., Christensen, D., Kankotia, R., Falk, I., Paxton, B.E., Kim, C.Y., 2013. Bacteriophage K for reduction of *Staphylococcus aureus* biofilm on central venous catheter material. Bacteriophage 3, e26825. https://doi.org/10.4161/bact.26825.
Luo, Y., McLean, D.T., Linden, G.J., McAuley, D.F., McMullan, R., Lundy, F.T., 2017. The naturally occurring host defense peptide, LL-37, and its truncated mimetics KE-18 and KR-12 have selected biocidal and antibiofilm activities against *Candida albicans*, *Staphylococcus aureus*, and *Escherichia* coli in vitro. Front. Microbiol. 8, 544. https://doi.org/10.3389/fmicb.2017.00544.
Mai, B., Wang, X., Liu, Q., Leung, A.W., Wang, X., Xu, C., Wang, P., 2016. The antibacterial effect of sinoporphyrin sodium photodynamic therapy on *Staphylococcus aureus* planktonic and biofilm cultures. Lasers Surg. Med. 48, 400–408. https://doi.org/10.1002/lsm.22468.
Mann, E.E., Rice, K.C., Boles, B.R., Endres, J.L., Ranjit, D., Chandramohan, L., Tsang, L.H., Smeltzer, M.S., Horswill, A.R., Bayles, K.W., 2009. Modulation of eDNA release and degradation affects *Staphylococcus aureus* biofilm maturation. PLoS One 4, e5822. https://doi.org/10.1371/journal.pone.0005822.
Manner, S., Goeres, D.M., Skogman, M., Vuorela, P., Fallarero, A., 2017. Prevention of *Staphylococcus aureus* biofilm formation by antibiotics in 96-microtiter well plates and drip flow reactors: critical factors influencing outcomes. Sci. Rep. 7, 43854. https://doi.org/10.1038/srep43854.

Mattila, S., Ruotsalainen, P., Jalasvuori, M., 2015. On-demand isolation of bacteriophages against drug-resistant bacteria for personalized phage therapy. Front. Microbiol. 6, 1271. https://doi.org/10.3389/fmicb.2015.01271.

Merino, N., Toledo-Arana, A., Vergara-Irigaray, M., Valle, J., Solano, C., Calvo, E., Lopez, J.A., Foster, T.J., Penadés, J.R., Lasa, I., 2009. Protein A-mediated multicellular behavior in Staphylococcus aureus. J. Bacteriol. 191, 832–843. https://doi.org/10.1128/JB.01222-08.

Mihu, M.R., Cabral, V., Pattabhi, R., Tar, M.T., Davies, K.P., Friedman, A.J., Martinez, L.R., Nosanchuk, J.D., 2017. Sustained nitric oxide-releasing nanoparticles interfere with methicillin resistant *Staphylococcus aureus* adhesion and biofilm formation in a rat central venous catheter model. Antimicrob. Agents Chemother. 61, e02020–16. https://doi.org/10.1128/AAC.02020-16.

Mishra, B., Golla, R.M., Lau, K., Lushnikova, T., Wang, 2015. Anti-staphylococcal biofilm effects of human cathelicidin peptides. G1ACS Med. Chem Lett. 7, 117–121. https://doi.org/10.1021/acsmedchemlett.5b00433.

Mohandas, A., Krishnan, A.G., Biswas, R., Menon, D., Nair, M.B., 2017. Antibacterial and cytocompatible nanotextured Ti surface incorporating silver via single step hydrothermal processing. Mater. Sci. Eng. C 75, 115–124. https://doi.org/10.1016/j.msec.2017.02.037.

Moncla, B.J., Pryke, K., Rohan, L.C., Graebing, P.W., 2011. Degradation of naturally occurring and engineered antimicrobial peptides by proteases. Adv. Biosci. Biotechnol. 2, 404–408.

Moormeier, D.E., Bose, J.L., Horswill, A.R., Bayles, K.W., 2014. Temporal and stochastic control of *Staphylococcus aureus* biofilm development. MBio. 5. https://doi.org/10.1128/mBio.01341-14. e01341-14.

Mootz, J.M., Malone, C.L., Shaw, L.N., Horswill, A.R., 2013. Staphopains modulate *Staphylococcus aureus* biofilm integrity. Infect. Immun. 81, 3227–3238. https://doi.org/10.1128/IAI.00377-13.

Morán, A., Gutiérrez, S., Martínez-Blanco, H., Ferrero, M.A., Monteagudo-Mera, A., Rodríguez-Aparicio, L.B., 2014. Non-toxic plant metabolites regulate *Staphylococcus* viability and biofilm formation: a natural therapeutic strategy useful in the treatment and prevention of skin infections. Biofouling 30, 1175–1182. https://doi.org/10.1080/08927014.2014.976207.

Mottola, C., Matias, C.S., Mendes, J.J., Melo-Cristino, J., Tavares, L., Cavaco-Silva, P., Oliveira, M., 2016. Susceptibility patterns of *Staphylococcus aureus* biofilms in diabetic foot infections. BMC Microbiol. 16, 119. https://doi.org/10.1186/s12866-016-0737-0.

Mu, H., Tang, J., Liu, Q., Sun, C., Wang, T., Duan, J., 2016. Potent antibacterial nanoparticles against biofilm and intracellular bacteria. Sci. Rep. 6, 18877. https://doi.org/10.1038/srep18877.

Nair, N., Vinod, V., Suresh, M.K., Vijayrajratnam, S., Biswas, L., Peethambaran, R., Vasudevan, A.K., Biswas, R., 2015. Amidase, a cell wall hydrolase, elicits protective immunity against *Staphylococcus aureus* and *S. epidermidis*. Int. J. Biol. Macromol. 77, 314–321. https://doi.org/10.1016/j.ijbiomac.2015.03.047.

Nair, S.V., Baranwal, G., Chatterjee, M., Sachu, A., Vasudevan, A.K., Bose, C., Banerji, A., Biswas, R., 2016. Antimicrobial activity of plumbagin, a naturally occurring naphthoquinone from Plumbago rosea, against *Staphylococcus aureus* and *Candida albicans*. Int. J. Med. Microbiol. 306, 237–248. https://doi.org/10.1016/j.ijmm.2016.05.004.

Nickerson, N., Ip, J., Passos, D.T., McGavin, M.J., 2010. Comparison of Staphopain A (ScpA) and B (SspB) precursor activation mechanisms reveals unique secretion kinetics of proSspB (Staphopain B), and a different interaction with its cognate Staphostatin. SspC Mol. Microbiol. 75, 161–177. https://doi.org/10.1111/j.1365-2958.2009.06974.x.

O'Neill, E., Pozzi, C., Houston, P., Humphreys, H., Robinson, D.A., Loughman, A., Foster, T.J., O'Gara, J.P., 2008. A novel *Staphylococcus aureus* biofilm phenotype mediated by the fibronectin-binding proteins, FnBPA and FnBPB. J. Bacteriol. 190, 3835–3850. https://doi.org/10.1128/JB.00167-08.

O'Neill, E., Humphreys, H., O'Gara, J.P., 2009. Carriage of both the fnbA and fnbB genes and growth at 37 °C promote FnBP-mediated biofilm development in meticillin-resistant *Staphylococcus aureus* clinical isolates. J. Med. Microbiol. 58, 399–402. https://doi.org/10.1099/jmm.0.005504-0.

Otto, M., 2008. Staphylococcal biofilms. Curr. Top. Microbiol. Immunol. 322, 207–228. https://doi.org/10.1007/978-3-540-75418-3_10.

Park, J.H., Ahn, M.Y., Kim, Y.C., Kim, S.A., Moon, Y.H., Ahn, S.G., Yoon, J.H., 2012. *In vitro* and *in vivo* antimicrobial effect of photodynamic therapy using a highly pure chlorin e6 against *Staphylococcus aureus* Xen29. Biol. Pharm. Bull. 35, 509–514. https://doi.org/10.1248/bpb.35.509.

Payne, D.E., Martin, N.R., Parzych, K.R., Rickard, A.H., Underwood, A., Boles, B.R., 2013. Tannic acid inhibits *Staphylococcus aureus* surface colonization in an isaA-dependent manner. Infect. Immun. 81, 496–504. https://doi.org/10.1128/IAI.00877-12.

Pelgrift, R.Y., Friedman, A.J., 2013. Nanotechnology as a therapeutic tool to combat microbial resistance. Adv. Drug Deliv. Rev. 65, 1803–1815. https://doi.org/10.1016/j.addr.2013.07.011.

Pérez-Laguna, V., Pérez-Artiaga, L., Lampaya-Pérez, V., García-Luque, I., Ballesta, S., Nonell, S., Paz-Cristobal, M.P., Gilaberte, Y., Rezusta, A., 2017. Bactericidal effect of photodynamic therapy, alone or in combination with mupirocin or linezolid, on Staphylococcus aureus. Front. Microbiol. 8, 1002. https://doi.org/10.3389/fmicb.2017.01002.

Pratiwi, S.U.T., Lagendijk, E.L., Hertiani, T., De Weert, S., Cornellius, A.M., Van Den Hondel, J.J., 2015. Antimicrobial effects of indonesian medicinal plants extracts on planktonic and biofilm growth of pseudomonas Aeruginosa and *Staphylococcus aureus*. Int. J. Pharm. Pharm. Sci. 7, 183–191. https://doi.org/10.4172/2376-0354.1000119.

Qi, X., Zhao, Y., Zhang, J., Han, D., Chen, C., Huang, Y., Chen, X., Zhang, X., Wang, T., Li, X., 2016. Increased effects of extracorporeal shock waves combined with Gentamicin against *Staphylococcus aureus* biofilms *in vitro* and *in vivo*. Ultrasound Med. Biol. 42, 2245–2252. https://doi.org/10.1016/j.ultrasmedbio.2016.04.018.

Qin, H., Cao, H., Zhao, Y., Zhu, C., Cheng, T., Wang, Q., Peng, X., Cheng, M., Wang, J., Jin, G., Jiang, Y., Zhang, X., Liu, X., Chu, P.K., 2014. *In vitro* and *in vivo* anti-biofilm effects of silver nanoparticles immobilized on titanium. Biomaterials 35, 9114–9125. https://doi.org/10.1016/j.biomaterials.2014.07.040.

Quave, C.L., Estévez-Carmona, M., Compadre, C.M., Hobby, G., Hendrickson, H., Beenken, K.E., Smeltzer, M.S., 2012. Ellagic acid derivatives from *Rubus ulmifolius* inhibit *Staphylococcus aureus* biofilm formation and improve response to antibiotics. PLoS One 7, e28737. https://doi.org/10.1371/journal.pone.0028737.

Renner, L.D., Weibel, D.B., 2011. Physicochemical regulation of biofilm formation. MRS Bull. 36, 347–355. https://doi.org/10.1557/mrs.2011.65.

Romero, C.M., Vivacqua, C.G., Abdulhamid, M.B., Baigori, M.D., Slanis, A.C., de Allori, M.C.G., Tereschuk, M.L., 2016. Biofilm inhibition activity of traditional medicinal plants from Northwestern Argentina against native pathogen and environmental microorganisms. Rev. Soc. Bras. Med. Trop. 49, 703–712. https://doi.org/10.1590/0037-8682-0452-2016.

Römling, U., Balsalobre, C., 2012. Biofilm infections, their resilience to therapy and innovative treatment strategies. J. Intern. Med. 272, 541–561. https://doi.org/10.1111/joim.12004.

Rosa, L.P., Da Silva, F.C., Nader, S.A., Meira, G.A., Viana, M.S., 2015. Antimicrobial photodynamic inactivation of *Staphylococcus aureus* biofilms in bone specimens using methylene blue, toluidine blue ortho and malachite green: an *in vitro* study. Arch. Oral Biol. 60, 675–680. https://doi.org/10.1016/j.archoralbio.2015.02.010.

Rozalski, M., Micota, B., Sadowska, B., Stochmal, A., Jedrejek, D., Wieckowska-Szakiel, M., Rozalska, B., 2013. Antiadherent and antibiofilm activity of *Humulus lupulus* L. derived products: new pharmacological properties. Biomed Res. Int., 101089. https://doi.org/10.1155/2013/101089. 2013.

Saginur, R., St. Denis, M., Ferris, W., Aaron, S.D., Chan, F., Lee, C., Ramotar, K., 2006. Multiple combination bactericidal testing of staphylococcal biofilms from implant-associated infections. Antimicrob. Agents Chemother. 50, 55–61. https://doi.org/10.1128/AAC.50.1.55-61.2006.

Sampath, L.A., Tambe, S.M., Modak, S.M., 2001. *In vitro* and *in vivo* efficacy of catheters impregnated with antiseptics or antibiotics: evaluation of the risk of bacterial resistance to the antimicrobials in the catheters. Infect. Control Hosp. Epidemiol. 22, 640–646. https://doi.org/10.1086/501836.

Sass, P., Bierbaum, G., 2007. Lytic activity of recombinant bacteriophage phi11 and phi12 endolysins on whole cells and biofilms of Staphylococcus aureus. Appl. Environ. Microbiol. 73, 347–352. https://doi.org/10.1128/AEM.01616-06.

Schaeffer, C.R., Woods, K.M., Longo, G.M., Kiedrowski, M.R., Paharik, A.E., Büttner, H., Christner, M., Boissy, R.J., Horswill, A.R., Rohde, H., Fey, P.D., 2015. Accumulation-associated protein enhances Staphylococcus epidermidis biofilm formation under dynamic conditions and is required for infection in a rat catheter model. Infect. Immun. 83, 214–226. https://doi.org/10.1128/IAI.02177-14.

Schwartz, K., Ganesan, M., Payne, D.E., Solomon, M.J., Boles, B.R., 2016. Extracellular DNA facilitates the formation of functional amyloids in *Staphylococcus aureus* biofilms. Mol. Microbiol. 99, 123–134. https://doi.org/10.1111/mmi.13219.

Sharlow, E.R., Leimgruber, S., Murray, S., Lira, A., Sciotti, R.J., Hickman, M., Hudson, T., Leed, S., Caridha, D., Barrios, A.M., Close, D., Grögl, M., Lazo, J.S., 2014. Auranofin is an apoptosis-simulating agent with in vitro and in vivo anti-leishmanial activity. ACS Chem. Biol. 9, 663–672. https://doi.org/10.1021/cb400800q.

Sharma, M., Visai, L., Bragheri, F., Cristiani, I., Gupta, P.K., Speziale, P., 2008. Toluidine blue-mediated photodynamic effects on staphylococcal biofilms. Antimicrob. Agents Chemother. 52, 299–305. https://doi.org/10.1128/AAC.00988-07.

Shaw, L., Golonka, E., Potempa, J., Foster, S.J., 2004. The role and regulation of the extracellular proteases of *Staphylococcus aureus*. Microbiology 150, 217–228. https://doi.org/10.1099/mic.0.26634-0.

Shirtliff, M.E., Mader, J.T., Camper, A.K., 2002. Molecular interactions in biofilms. Chem. Biol. 9, 859–871. https://doi.org/10.1016/S1074-5521(02)00198-9.

Singh, R., Lillard, J.W., 2009. Nanoparticle-based targeted drug delivery. Exp. Mol. Pathol. https://doi.org/10.1016/j.yexmp.2008.12.004.

Singh, A.V., Vyas, V., Patil, R., Sharma, V., Scopelliti, P.E., Bongiorno, G., Podestà, A., Lenardi, C., Gade, W.N., Milani, P., 2011. Quantitative characterization of the influence of the nanoscale morphology of nanostructured surfaces on bacterial adhesion and biofilm formation. PLoS One 6. https://doi.org/10.1371/journal.pone.0025029.

Singh, R., Sahore, S., Kaur, P., Rani, A., Ray, P., 2016. Penetration barrier contributes to bacterial biofilm-associated resistance against only select antibiotics, and exhibits genus-, strain- and antibiotic-specific differences. Pathog. Dis. 74. https://doi.org/10.1093/femspd/ftw056.

Skogman, M.E., Kujala, J., Busygin, I., Leinob, R., Vuorela, P.M., Fallarero, A., 2012. Evaluation of antibacterial and anti-biofilm activities of cinchona alkaloid derivatives against Staphylococcus aureus. Nat. Prod. Commun. 7, 1173–1176.

Speziale, P., Pietrocola, G., Foster, T.J., Geoghegan, J.A., 2014. Protein-based biofilm matrices in Staphylococci. Front. Cell. Infect. Microbiol. 4. https://doi.org/10.3389/fcimb.2014.00171.

Sugimoto, S., Sato, F., Miyakawa, R., Chiba, A., Onodera, S., Hori, S., Mizunoe, Y., 2018. Broad impact of extracellular DNA on biofilm formation by clinically isolated Methicillin-resistant and -sensitive strains of *Staphylococcus aureus*. Sci. Rep. https://doi.org/10.1038/s41598-018-20485-z.

Thammavongsa, V., Kim, H.K., Missiakas, D., Schneewind, O., 2015. Staphylococcal manipulation of host immune responses. Nat. Rev. Microbiol. https://doi.org/10.1038/nrmicro3521.

Thangamani, S., Mohammad, H., Abushahba, M.F.N., Sobreira, T.J.P., Seleem, M.N., 2016. Repurposing auranofin for the treatment of cutaneous staphylococcal infections. Int. J. Antimicrob. Agents 47, 195–201. https://doi.org/10.1016/j.ijantimicag.2015.12.016.

Thurlow, L.R., Hanke, M.L., Fritz, T., Angle, A., Aldrich, A., Williams, S.H., Engebretsen, I.L., Bayles, K.W., Horswill, A.R., Kielian, T., 2011. *Staphylococcus aureus* biofilms prevent macrophage phagocytosis and attenuate inflammation in vivo. J. Immunol. 186, 6585–6596. https://doi.org/10.4049/jimmunol.1002794.

Toledo-Arana, A., Merino, N., Vergara-Irigaray, M., Débarbouillé, M., Penadés, J.R., Lasa, I., 2005. *Staphylococcus aureus* develops an alternative, ica-independent biofilm in the absence of the arlRS two-component system. J. Bacteriol. 187, 5318–5329. https://doi.org/10.1128/JB.187.15.5318-5329.2005.

Tong, S.Y.C., Davis, J.S., Eichenberger, E., Holland, T.L., Fowler, V.G., 2015. *Staphylococcus aureus* infections: epidemiology, pathophysiology, clinical manifestations, and management. Clin. Microbiol. Rev. 28, 603–661. https://doi.org/10.1128/CMR.00134-14.

Torres, N.S., Abercrombie, J.J., Srinivasan, A., Lopez-Ribot, J.L., Ramasubramanian, A.K., Leung, K.P., 2016. Screening a commercial library of pharmacologically active small molecules against *Staphylococcus aureus* biofilms. Antimicrob. Agents Chemother. 60, 5663–5672. https://doi.org/10.1128/AAC.00377-16.

Tran, P.L., Lowry, N., Campbell, T., Reid, T.W., Webster, D.R., Tobin, E., Aslani, A., Mosley, T., Dertien, J., Colmer-Hamood, J.A., Hamood, A.N., 2012. An organoselenium compound inhibits *Staphylococcus aureus* biofilms on hemodialysis catheters in vivo. Antimicrob. Agents Chemother. https://doi.org/10.1128/AAC.05680-11.

Van den Driessche, F., Brackman, G., Swimberghe, R., Rigole, P., Coenye, T., 2017. Screening a repurposing library for potentiators of antibiotics against *Staphylococcus aureus* biofilms. Int. J. Antimicrob. Agents 49, 315–320. https://doi.org/10.1016/j.ijantimicag.2016.11.023.

Van Praagh, A.D.G., Li, T., Zhang, S., Arya, A., Chen, L., Zhang, X.X., Bertolami, S., Mortin, L.I., 2011. Daptomycin antibiotic lock therapy in a rat model of staphylococcal Central venous catheter biofilm infections. Antimicrob. Agents Chemother. https://doi.org/10.1128/AAC.00147-11.

Vergara-Irigaray, M., Valle, J., Merino, N., Latasa, C., García, B., De Los Mozos, I.R., Solano, C., Toledo-Arana, A., Penadés, J.R., Lasa, I., 2009. Relevant role of fibronectin-binding proteins in *Staphylococcus aureus* biofilm-associated foreign-body infections. Infect. Immun. 77, 3978–3991. https://doi.org/10.1128/IAI.00616-09.

Walencka, E., Sadowska, B., Rózalska, S., Hryniewicz, W., Rózalska, B., 2005. Lysostaphin as a potential therapeutic agent for staphylococcal biofilm eradication. Polish J. Microbiol. 54, 191–200.

Weigel, L.M., Donlan, R.M., Shin, D.H., Jensen, B., Clark, N.C., McDougal, L.K., Zhu, W., Musser, K.A., Thompson, J., Kohlerschmidt, D., Dumas, N., Limberger, R.J., Patel, J.B., 2007. High-level vancomycin-resistant *Staphylococcus aureus* isolates associated with a polymicrobial biofilm. Antimicrob. Agents Chemother. 51, 231–238. https://doi.org/10.1128/AAC.00576-06.

Wen, H., Dan, M., Yang, Y., Lyu, J., Shao, A., Cheng, X., Chen, L., Xu, L., 2017. Acute toxicity and genotoxicity of silver nanoparticle in rats. PLoS One 12. https://doi.org/10.1371/journal.pone.0185554.

Wood, T.K., Knabel, S.J., Kwan, B.W., 2013. Bacterial persister cell formation and dormancy. Appl. Environ. Microbiol. https://doi.org/10.1128/AEM.02636-13.

Wu, J.A., Kusuma, C., Mond, J.J., Kokai-Kun, J.F., 2003. Lysostaphin disrupts *Staphylococcus aureus* and Staphylococcus epidermidis biofilms on artificial surfaces. Antimicrob. Agents Chemother. 47, 3407–3414. https://doi.org/10.1128/AAC.47.11.3407-3414.2003.

Wu, X., Wang, Y., Tao, L., 2011. Sulfhydryl compounds reduce *Staphylococcus aureus* biofilm formation by inhibiting PIA biosynthesis. FEMS Microbiol. Lett. https://doi.org/10.1111/j.1574-6968.2010.02190.x.

Xiong, Y.Q., Estellés, A., Li, L., Abdelhady, W., Gonzales, R., Bayer, A.S., Tenorio, E., Leighton, A., Ryser, S., Kauvar, L.M., 2017. A human biofilm-disrupting monoclonal antibody potentiates antibiotic efficacy in rodent models of both *Staphylococcus aureus* and Acinetobacter baumannii infections. Antimicrob. Agents Chemother. 61. https://doi.org/10.1128/AAC.00904-17.

Yu, Z., Deslouches, B., Walton, W.G., Redinbo, M.R., Di, Y.P., 2018. Enhanced biofilm prevention activity of a SPLUNC1-derived antimicrobial peptide against Staphylococcus aureus. PLoS One 13. https://doi.org/10.1371/journal.pone.0203621.

Zapotoczna, M., Forde, É., Hogan, S., Humphreys, H., O'Gara, J.P., Fitzgerald-Hughes, D., Devocelle, M., O'Neill, E., 2017. Eradication of *Staphylococcus aureus* biofilm infections using synthetic antimicrobial peptides. J. Infect. Dis. 215, 975–983. https://doi.org/10.1093/infdis/jix062.

Zhang, Q.Z., Zhao, K.Q., Wu, Y., Li, X.H., Yang, C., Guo, L.M., Liu, C.H., Qu, D., Zheng, C.Q., 2017. 5-aminolevulinic acid-mediated photodynamic therapy and its straindependent combined effect with antibiotics on *Staphylococcus aureus* biofilm. PLoS One 12. https://doi.org/10.1371/journal.pone.0174627.

Zhao, X., Wu, H., Lu, H., Li, G., Huang, Q., 2013. Lamp: a database linking antimicrobial peptides. PLoS One 8. https://doi.org/10.1371/journal.pone.0066557.

Zhou, Y., Zhao, R., Ma, B., Gao, H., Xue, X., Qu, D., Li, M., Meng, J., Luo, X., Hou, Z., 2016. Oligomerization of RNAIII-Inhibiting peptide inhibits adherence and biofilm formation of methicillin-resistant *Staphylococcus aureus* in vitro and in vivo. Microb. Drug Resist. 22, 193–201. https://doi.org/10.1089/mdr.2015.0170.