Genij Ortopedii. 2021. Vol. 27, no. 5. P. 628-635.

Review article

https://doi.org/10.18019/1028-4427-2021-27-5-628-635

Pathogenetic features of chronic osteomyelitis treatment

D.V. Rimashevskiy[1]✉, I.F. Akhtyamov[2], P.N. Fedulichev[3], Wessam Zaalan[1], K.A. Ustazov[1], Abdul Basith[1], J.M. Moldakulov[4], M.P. Zinoviev[5]

[1]Peoples' Friendship University of Russia, Moscow, Russian Federation
[2]Kazan State Medical University, Kazan, Russian Federation
[3]S.I. Georgievsky Medical Academy – V.I. Vernadsky Crimean Federal University, Simferopol, Russian Federation
[4]Taldykorgan city multidisciplinary hospital, Taldykorgan, Republic of Kazakhstan
[5]Federal Center for Traumatology, Orthopedics and Endoprosthetics, Smolensk, Russian Federation

Corresponding author: Denis V. Rimashevskiy, drimashe@yandex.ru

Abstract

Over the past decades, there has been a steady increase in the incidence of osteomyelitis. It is associated with an increased use of implants in traumatology and orthopedics. The social aspects of osteomyelitis are, on the one hand, significant financial costs for the healthcare system, and on the other hand, high recurrence and re-infection in the treatment of joint pathology associated with long-term loss of work ability and a high risk of patient's disability. **Purpose** To conduct a search and analysis of publications in Russian and English, devoted to the problem of osteomyelitis and periprosthetic infection, on the basis of which to summarize the main current notions about the etiology, pathogenesis, diagnosis and treatment of osteomyelitis. **Materials and methods** The search was carried out in the Pubmed and CyberLeninka databases of literature sources over the past 10 years. The data were analyzed and compared with the materials from earlier publications. Only publications from peer-reviewed journals were considered for analysis. **Results and discussion** Success in the treatment of peri-implant infection with prosthesis re-implantation and satisfactory joint function has been achieved in only just more than a half of patients. Recent studies have significantly changed the understanding of the etiology and pathogenesis of osteomyelitis. It has been proven that in osteomyelitis and implant-associated infection, four reservoirs of infection are formed in the patient's body: abscesses in soft tissues and bone marrow canal, biofilms on the surface of implants and necrotic tissues, intracellular colonization with bacteria of the macroorganism and lacunar-canalicular system. Understanding the mechanisms of osteomyelitis development and its course forces the specialists to take a fresh look at the causes of failures in the fight against such a severe pathology and change approaches to its prevention, diagnosis and treatment.

Keywords: osteomyelitis, periprosthetic infection, biofilm, etiology, pathogenesis, staphylococcus

For citation: Rimashevskiy D.V., Akhtyamov I.F., Fedulichev P.N., Zaalan Wessam, Ustazov K.A., Basith Abdul, Moldakulov J.M., Zinoviev M.P. Pathogenetic features of chronic osteomyelitis treatment (literature review) *Genij Ortopedii*, 2021, vol. 27, no 5, pp. 628-635. https://doi.org/10.18019/1028-4427-2021-27-5-628-635

INTRODUCTION

Over the past 50 years, there has been a steady increase in the incidence of osteomyelitis and periprosthetic infection (PPI). It is associated with an increased the number of implants used and a growing number of people with diabetes mellitus [1]. The frequency of revision interventions for PPI after primary total knee arthroplasty (TKA) is 7.5 cases per 1000 joints [2]. Revision TKA in PPI is a complex operation and is associated with a high incidence of postoperative complications. The five-year mortality rate of patients with PPI is comparable to that of some common types of cancer, and more than 15 % of patients require re-revision by 10 years [2, 3]. The social significance of osteomyelitis is not limited to the large financial costs for the health care system. Thus, a recent study in Taiwan showed that the risk of suicide among patients with chronic osteomyelitis is almost 2 times higher than the average in the population [4].

Despite aggressive surgical tactics, including radical surgery with removal or replacement of all implants and prolonged antibiotic therapy, the infection recurs. Failures or re-infection in the surgical treatment of osteomyelitis and PPI reaches 33 % [2, 5], and a successful outcome according to the MSIS RT (Musculoskeletal Infection Society Outcome Reporting Tool) criteria is achieved only in 55 % of PPI cases in knee and hip arthroplasty [6]. The complexity, long duration and high cost of osteomyelitis treatment, high incidence of unfavorable outcomes made this disease one of the priority areas in the search for clinical and cost-effective methods of prevention and treatment [2].

The **aim** of the work was to search and analyze publications in Russian and English, devoted to the problem of osteomyelitis and periprosthetic infection, and to present a brief review of the main modern concepts about the etiology, pathogenesis, diagnosis and treatment of osteomyelitis.

MATERIAL AND METHODS

The Pubmed and CyberLeninka databases were searched for literary sources published in the past 10 years. The data were analyzed and compared with the materials from earlier publications. Only publications from peer-reviewed journals were considered.

RESULTS

Etiopathogenesis

Most cases of osteomyelitis and periprosthetic infection (PPI) are associated with staphylococcus [7, 8]. In a number of clinics, more than 50 % of PPI cases have been caused by methicillin-resistant *S. aureus* (MRSA) strains [11]. Osteomyelitis can also be caused by enterococci, pseudomonas, streptococci and other microorganisms [9]. A big threat is the growing prevalence of osteomyelitis caused by *Acinetobacter baumannii*, which is extremely resistant to most antibacterial drugs [10, 11]. In chronic osteomyelitis, the infection can be either monomicrobial (62–81 %) or polymicrobial (11–38 %) [12].

Pathogenesis of osteomyelitis caused by *S. aureus* has been studied best of all. *S. aureus* is an extremely versatile opportunistic microorganism capable of infecting almost all organs and systems in the human body and causing life-threatening diseases [14]. Moreover, 20–60 % of people colonized with *S. aureus* are asymptomatic [14]. It possesses a number of virulence factors and resistance mechanisms, including the secretion of toxins [15], the ability to adhere as a way to evade the immune response [16], form biofilms [17] and small colony variants with slow growth (SCV) [18], and to develop antimicrobial resistance [19]. As a result of these complex pathogenetic defense mechanisms against the host immune system, recurrence of *S. aureus* osteomyelitis may occur after decades of clinical latency [20].

A retrospective analysis of 825 one-stage revisions for PPI after total hip arthroplasty by Buchholz et al. in 1984 showed that *S. aureus* was the most common pathogen, and the 5-year survival rate after revision for recurrence of infection was only 77 % [21]. It is noteworthy that the results of the international consensus meeting on infections of the musculoskeletal system in 2018 showed that the current results in PPI rates do not differ from the results fifty years ago, type of primary pathogen, treatment algorithm and the proportion of negative treatment outcomes [22].

Recent studies show that the body has four pathogenetically distinct reservoirs of bacteria in osteomyelitis, and each of them must be treated, otherwise the infection will persist or recur [23].

Bone and soft tissue abscesses

Abscesses are most often observed in the skin, but may develop in any tissue after dissemination of microbes from the primary nidus [24]. Abscess formation is a host defense reaction, a dynamic process controlled not only by the host signals, but also by the pathogen signals [24]. The formation of an abscess begins with the migration of neutrophils to the nidus of acute infection, stimulated by the local release of cytokines and chemokines by the host cells. Activated neutrophils begin to

ht extracellular bacteria by phagocytosis, degranulation, and formation of neutrophilic extracellular traps (NETosis) [25]. *S. aureus* is capable of attacking neutrophils and other phagocytes, releasing cytolytic toxins that create pores in the host cell membranes [26]. Abscesses may be detected as early as 4 days after infection [26]. The abscess center consists of living bacteria surrounded by viable and necrotic polymorphonuclear leukocytes separated from healthy tissue by a layer of fibrin lined with macrophages designated to prevent bacterial dissemination [27]. As the abscess matures over several weeks, the immune cells remain only in the periphery, where they cannot access bacteria, which in turn continue to multiply in the center of the abscess. Our immune system cannot cope with abscesses on its own. Without outside intervention, persistence within the abscess can lead to rupture of its membrane and dissemination of bacteria [24]. Thus, success in the treatment of infections, especially implant-associated infections, largely depends on thorough surgical debridement of soft tissues and bone and the adequacy of subsequent local antimicrobial therapy to eliminate the remaining in the wound bacteria.

Biofilms on implants

Implant-associated osteomyelitis due to *S. aureus* is initiated with adhesion of bacterial cells to extracellular matrix components known Microbial Surface Components Recognizing Adhesive Matrix Molecules (MSCRAMMs) [25]. As the acronym suggests, MSCRAMMs are proteins on the surface of microbial cells capable of binding extracellular matrix ligands such as fibronectin, fibrinogen, and collagen. The adhesion of bacterial cells to the substrate creates conditions for subsequent local proliferation and colonization. The host's immune response to bacterial colonization begins with the release of a number of proinflammatory cytokines, chemokines, antimicrobial peptides by innate immune cells, which triggers the migration of neutrophils to the site of infection. At this time, neutrophils and other cells of innate immunity bind free (planktonic) bacterial cells and try to destroy them. At some point, the PMNL attack dies down when all planktonic bacteria are destroyed, or they are hidden in biofilms. Biofilm formation can be divided into four stages: (1) bacterial cell attachment, (2) proliferation, (3) biofilm maturation, and (4) detachment and dissemination. As the biofilm matures, it becomes a complex, heterogeneous structure with bacterial cells, empty spaces, and complex channels to facilitate the transport of nutrients and oxygen through the bulk of the biofilm. Finally, detachment or dispersal of biofilm allows bacterial cells to metastasize in the fluid flow to new and non-colonized regions of the host [28].

The sense of quorum ("quorum sensing") is a communication system between cells that controls the expression of many genes in response to changes

in population density [29]. Thanks to this system, intercellular information exchange is carried out to facilitate the dynamic expression of a number of virulence genes, including those responsible for biofilm formation [30]. Biofilm ensures long-term survival of bacterial cells in a hostile environment through various mechanisms. First, the biofilm protects bacterial cells from the immune system by providing a physical barrier to the entry of immune cells, protecting against phagocytosis and death from reactive oxygen species (ROS). In the biofilm, bacteria are able to change their phenotype and acquire resistance to antimicrobial drugs. *S. aureus* is able to alter the growth rate, dependence on oxygen and nutrients, and exhibit new virulence mechanisms acquired through horizontal gene transfer [31]. All this makes the bacteria inside biofilms extremely resistant to antimicrobial therapy. The minimum inhibitory concentration (MIC) of antibiotics for microbes in the biofilm may be thousands times higher than the MIC for planktonic forms [32]. In addition to ensuring the long-term survival of bacteria, biofilms are able to greatly damage the surrounding host tissues. Bacterial infections cause bone resorption both indirectly through host inflammatory factors and directly by bacterial factors [33, 34].

Thus, the biofilm is a significant problem for surgeons in implant-associated infection. It is obvious that radical removal of the biofilm from implants and surrounding necrotic tissues is of fundamental importance for arresting the infectious process. Unfortunately, it is not possible to completely remove the mature biofilm from implants by washing or ultrasound [35]. Therefore, radical surgical debridement and removal of all implants and bone cement residues has been considered the gold standard to reduce the risk of reinfection.

Colonization of the lacunar-canalicular system (LCS)

The third and only recently discovered reservoir of bacteria in chronic staphylococcal osteomyelitis is the colonization of LCS [36]. Transmission electron microscopy (TEM) of the affected bone showed *S. aureus* invasion into the tubular canaliculi perpendicular to the medullary canal and subsequent colonization of lacunar spaces devoid of osteocytes.

It has been proven that colonization of LCS by *S. aureus* is an active process [36]. Lacking any visible motility structures such as flagella, cilia, or pseudopodia, it is assumed that *S. aureus* is able to enter the canaliculi of living bone using a novel motility mechanism. It is believed that LCS invasion begins with the death of cells of the cortical bone endosteum due to local inflammation at the initial stage of infection with *S. aureus*, which can find open bone canaliculi and submerge there deformed daughter cells by asymmetric division [37]. Colonized canaliculi are often widened and scalloped as the bacterial acid demineralizes bone and the bacteria consume organic bone matrix. This process is also observed in ex vivo studies regardless of host factors [34]. The invasion proceeds with the proliferation of *S. aureus* cells in the canaliculi and lacunar spaces, since osteocytes die as a result of bacterial colonization. Unlike bacteria in the bone marrow, which are accessible to neutrophils, *S. aureus* inside the LCS is surrounded by a dense mineral matrix of the cortical bone and is completely inaccessible to immune cells. These observations suggest that *S. aureus* is able to survive for decades within the LCS with an inexhaustible supply of nutrients, while avoiding attacks by the immune system [38].

The discovery of the LCS invasion has showen that if, when performing necrectomy, one is guided solely by the appearance of the bone ("white" – dead, "red" – alive), one cannot be completely sure of accurate removal of the infected bone. Bone colonized by *S. aureus* may remain and contribute to the recurrence of infection. In a mouse model of osteomyelitis, deep bacterial invasion of the cortical bone occurred in the first two weeks. This suggests that this time interval separates acute infection from chronic infection [38]. **The very possibility of eradication of bacteria infecting LCS is questioned, since the efficacy of standard antibacterial therapy against *S. aureus* embedded in the bone matrix has been currently unknown. Therefore, we can never say that our treatment achieved complete eradication of the infection in osteomyelitis or PPI, we can only state the sedation of infection**. In addition, it is quite possible that bacteria inside the LCS exhibit an altered growth phenotype similar to the phenotype of small colonies, which makes them resistant to antimicrobial therapy [39]. Libraty D.H. et al. described a particularly interesting case of osteomyelitis recurrence due to *S. aureus* 75 years after the patient was surgically treated without antibiotic therapy. Genetic analysis of the infectious agent showed that *S. aureus* belonged to the same strain that had been circulating in the patient's region of residence 75 years ago and retained sensitivity to penicillin and oxacillin [40].

An unresolved clinical task is a **diagnostic procedure that could determine the limits of bone infection and assess the completeness of surgical debridement during sanitizing operations**.

Intracellular invasion of *S. aureus*

In addition to the mechanisms of osteomyelitis persistence described above, there is ongoing debate over the importance of colonization in bone cell as a possible mechanism for chronic osteomyelitis [41]. Intracellular persistence of *S. aureus* in various types of human cells has been described many times [41]. It has been proven that *S. aureus* is able to survive in professional and non-professional phagocytes, including macrophages, neutrophils, fibroblasts, keratinocytes, epithelial and endothelial cells [42, 43]. Intracellular persistence of *S. aureus* in

osteoblasts and osteocytes is of particular concern due to the long lifespan of these cells [44]. In vitro studies have shown the ability of osteoclasts [45] and osteocytes [46] to internalize *S. aureus*. It should be noted that intracellularly infected leukocytes are often found in sepsis [47]. A particular concern is the possibility of intracellular persistence in leukocytes and macrophages due to the longer lifespan of these leukocytes and their ability to move in the circulatory system [48]. Zhu and co-authors have shown that intestinal neutrophils may act as the "Trojan horse" [48]. The authors colonized the rat intestines with MRSA eight or 72 hours before implanting the wire in the knee joint. The incidence of PPI ranged from 25 % to 35 %, depending on the MRSA strain. In order to exclude the influence of the colonization of the wound from the outside, a solution with MRSA was daily dripped onto the wound for 10 days after the operation in the control group of rats. To exclude the effect of bacteremia, another control group was injected intravenously with 10^8 CFU MRSA for 3 days after the operation. There were no cases of PPI in the control groups. At 1, 3, 5, and 7 days after intestine colonization, 15.3 %, 7.5 %, 5.6 %, and 5.1 % of circulating neutrophils were MRSA positive. Intravenous administration of colonized intestinal neutrophils (10^6) within 5 days after surgery resulted in PPI in 55 % of animals. The authors found that bacteria also enter neutrophils by intravenous administration of MRSA, but the amount of MRSA in blood neutrophils is only 1 % of their number in intestinal neutrophils [48].

In about 10 % of cases, PPI leads to sepsis and death, and *S. aureus* remains the leading and most lethal cause of bacteremia [49]. In a postmortem study of patients who died due to *S. aureus* infection from septic multiple organ failure, both extracellular bacteria and intracellularly colonized leukocytes were found. Live *S. aureus* in the cytoplasm of macrophages turn them into "Trojan horses". In a study directly comparing osteoblastic versus macrophage infection with *S. aureus*, the researchers found that macrophages contain 100 times more living bacteria than osteoblasts, and that osteoblasts are significantly less viable [50].

It is possible that intracellular infection plays a certain role in bone infection, but this role is not evident due to a limited in vivo evidence base. **Prescribing antibiotic therapy, one must remember about the possible intracellular colonization and use antibiotics with proven activity against intracellular microorganisms**.

Staphylococcus aureus has developed numerous mechanisms to effectively counteract the adaptive defenses of the human body. Anti- *S. aureus* antibodies are found in all people. We are in contact with *S. aureus* throughout our lives, what is manifested by acute infections or asymptomatic carriage [51, 52]. However, the presence of antibodies to *S. aureus* in the host (collectively referred to as humoral "immune proteomes") does not necessarily guarantee protection against this infection in the future. In fact, some individuals with antibodies to *S. aureus* are even more at risk of developing *S. aureus* PPI than others, probably because of the susceptible rather than defensive nature of their immune proteome [29]. It turned out that antibodies against some antigens (autolysins: Amd, Gmd; secreted immunotoxins: CHIPS, SCIN, Hla) are associated with protection against staphylococcal infection, while high levels of antibodies against iron-regulated surface determinant proteins (Isd: IsdA, IsdB, IsdH), on the contrary, are associated with unfavorable clinical outcomes [53]. **A better understanding of the functional role of specific antibodies in *S. aureus* infections may help predict outcomes and provide targets for the development of new therapies for this disease**.

Diagnosis

Chronic infections, such as implant-associated osteomyelitis, can present diagnostic difficulties because patients usually present with nonspecific symptoms, including pain, joint swelling, and fever.

Radiography plays a vital role in the diagnosis of osteomyelitis and can be used to assess the extension of infection and even search for specific signs that distinguish the acute stage of the disease from the chronic one, in which a fistulous tract can be found by performing fistulography (radiography with the introduction of contrast into the fistulous tract), sequestration (avascular necrotic bone) or the area of bone sclerosis around the lesion [54]. The diagnostic value of radiography is limited in low disease activity and presence of metal implants.

MRI is usually performed after plain radiography in patients with suspected osteomyelitis [55]. MRI enables to determine the spatial distribution of the process in soft tissue infection. It is believed that a normal picture of the bone marrow on MRI excludes osteomyelitis with 100 % probability [56]. However, the specificity of MRI in the diagnosis of osteomyelitis is limited to 83 %, i.e. the probability of a false positive result is 17 % [57]. The standard for MRI diagnosis of osteomyelitis is the use of gadolinium-based contrast, which is well visualized on T1 weighted images. But the use of gadolinium in patients with chronic renal failure is dangerous due to development of nephrogenic systemic fibrosis [58]. Optimistic results were shown by the use of "ferumoxytol", a drug originally created for the treatment of iron deficiency anemia. After administration of the drug, particles of supermagnetic iron oxide are slowly absorbed by cells of the immune system, mainly macrophages, and are well defined on T2 weighted images for several days. Unlike ferumoxytol, gadolinium gives the maximum signal shortly after administration and quickly leaves the tissues [59].

The sequestration is best visualized with CT, where it looks like a mineralized bone segment with a peripheral enlightenment zone. However, detection of sequestration on CT scans is not pathognomonic for chronic osteomyelitis. Several primary bone tumors can form a sequester-like mineralized matrix, especially osteoid tumors such as osteoid osteoma and osteoblastomas [60]. Unlike sequestration, which usually has rough edges, the calcification zone in the center of the osteoid – osteoma is smooth and round [61].

Blood cultures in most cases confirm the diagnosis of osteomyelitis of the spine [62] and should be performed immediately if there is clinical or radiological suspicion, but the probability of a false negative is 20 % for gram-positive flora and 50 % for gram-negative flora. The biopsy is more accurate and should be performed immediately if culture is negative [62].

Intraoperative histopathological examination may reveal areas of acute infection in periprosthetic tissue samples. The greatest value in such cases is the study of the membrane formed at the interface between the bone and the implant [63]. It was previously thought that acute inflammation, defined by the presence of > 1–10 PMNLs in several high-resolution visual fields (400 ×), may indicate an acute infection [64]. However, the variability of the manifestations of osteomyelitis is observed not only between patients, but also at the cellular level within the same lesion [65]. A definitive diagnosis of infection requires intraoperative samples or biopsies.

Most clinicians rely on inflammatory cell counts and markers such as C-reactive protein (CRP) and erythrocyte sedimentation rate (ESR) to diagnose orthopedic infections. However, these methods are neither pathogen-specific nor anatomically specific and do not accurately assess the response to therapy. Methods for diagnosing bone infection based on the assessment of the humoral immune response have been actively developing, but they have not received wide clinical use yet.

It is advisable to take materials for microbiological study at least two weeks after the withdrawal of antibiotics and to suspend preoperative antibiotic prophylaxis before harvesting the material. At least 5 deep tissue or fluid samples should be taken from the area of suspected infection adjacent to the fracture / implant. Samples of superficial tissue as well as fluids and fistula discharge should not be used to identify bacteria. The use of smears to determine microflora in osteomyelitis is unacceptable due to the low sensitivity of the method [66].

Treatment

In the 1970s, a standard was developed for the treatment of implant-associated osteomyelitis, and, first of all, periprosthetic infection (PPI), which includes: (1) removal of the infected implant; (2) extensive surgical debridement of the adjacent bone and soft tissues; (3) filling bone defects with antibiotic-loaded acrylic cement.

Treatment of osteomyelitis and peri-implant infection must be pathogenic. Before using antibiotics, it is necessary to collect material for microbiological tests. In chronic osteomyelitis and peri-implant infection, the clinician usually has a few days to more carefully analyze the causes of osteomyelitis and previous treatment failure. Modifiable risk factors for treatment failure must be eliminated [67]. For example, patients with HIV infection should receive antiviral therapy aimed at restoring the level of T-cells (CD-4); it is necessary to normalize and stabilize blood sugar levels, assess the state of major vessels and, if needed, discuss with a vascular surgeon the options for restoring blood flow in the affected limb in patients with diabetes mellitus [68]. It is necessary to demand that patients quit smoking, especially if reconstructive surgery on soft tissues with flap transfer is planned [67, 68].

The patient's clinical characteristics, age and microbiological profile of the geographic region should be taken into account by defining the diagnosis and choosing antibiotic therapy. It is known that *S. aureus* is more often detected in osteomyelitis of the spine in young patients, while in elderly patients gram-negative flora and enterococcus are more frequent. Gram-negative flora is twice as common in women than in men (32.1 % versus 16.4 %; $p < 0.05$), in patients with liver cirrhosis (32.7 % versus 21.1 % without cirrhosis; $p < 0.05$) and among patients with solid tumors (31.0 % versus 20.7 % without tumors; $p < 0.05$). MRSA is found more often in patients with chronic kidney disease (34.4 % versus 14.7 % in cases without chronic kidney disease; $p < 0.05$) [69]. This must be borne in mind when prescribing empiric antibiotic therapy in patients with osteomyelitis and negative culture results.

Antibiotics used in clinical practice either inhibit cell wall biosynthesis, protein, DNA, or RNA synthesis, or interfere with critical metabolic synthesis [70]. Recently, more and more attention as a target for antibacterial action is given to riboswitches, RNA regulatory elements located mainly in the 5'UTR of bacterial mRNA. Targeting riboswitches is a novel approach that is effective in combating drug-resistant bacteria and biofilm-associated infections [71]. One of these drugs, PKZ18-22, in an in-vitro study was 10 times more potent than vancomycin in inhibiting the growth of *S. aureus* in the biofilm and demonstrated synergism with existing antibiotics such as gentamicin and rifampicin [72].

It is known that aspirin effectively suppresses the "sense of quorum" in *P. aeruginosa* [73], is able to reduce the periosteal response, osteolysis, activation of osteoclasts, and activate osteoblasts in osteomyelitis [74]. In a recent study, it was demonstrated that administration of aspirin at a dosage of 100 mg/day in patients with PPI after TKA and THA had a beneficial effect on the

effectiveness of infection treatment [75]. Cannabinoids inhibit the biofilm-forming ability of MRSA and destroy the already-formed biofilm and dormant antibiotic-resistant stationary phase cells.

Cannabigerol acts on the cytoplasmic membrane of gram-positive bacteria and has been effective in vivo in systemic MRSA infection. Cannabinoids are effective against gram-negative organisms, the outer membrane of which is permeable, and cannabigerol acts on the inner membrane. The combination of cannabinoids with polymyxin B is effective against multidrug resistance of gram-negative pathogens and unlocks a wide spectrum of the therapeutic potential of cannabinoids.

The use of nanoparticles as carriers of antibiotics is of interest. Thus, it has been proven that the use of rifampicin and linezolid in lipid-polymer hybrid nanoparticles increases their effectiveness against intracellular MRSA by several times [76].

Immunotherapy can be an effective adjunct to antibiotics for treating intractable osteomyelitis. Vaccines have been developed over the past two decades for numerous potential targets of *S. aureus*, but none have been able to provide protection against *S. aureus* in humans. [77]. Passive immunization using monoclonal antibodies (mAb) is becoming increasingly attractive for the treatment of osteomyelitis caused by *S. aureus*. The advantage of passive immunization with mAbs is their high antigen-neutralizing specificity. In addition, the mAbs may be administered locally at the site of infection and the scale-up production of mAbs is not extremely expensive. Based on the clinical success of biological cancer immunotherapy with checkpoint inhibitors, it is likely that antibody-based biologics offer similar promise for the treatment of osteomyelitis. It is encouraging that some anti-*S. aureus* mAbs have been currently under preclinical and clinical studies [29]. Thus, Masters et al. proved that mAbs to penicillin-binding protein 4 type MRSA not only block the penetration of staphylococcus into submicron spaces, but also increase the effectiveness of antibiotic therapy by several times [43].

The choice of an antibiotic (AB) for mixing in a bone-cement spacer is limited by the sensitivity of the microflora, the ability of AB to withstand high temperatures during polymerization, and the duration of the half-life [78]. The release of ABs from the bone cement actively occurs only in the first days after implantation, subsequently it sharply decreases, and ABs continue to be released for a long time at a concentration below the MIC, which raises concerns about the development of flora resistance and recurrence of infection. Exposure to AB at a dose below the MIC may trigger the resistance in bacteria and small colonies (SCVs) with recurrent infection [78, 79].

In any treatment option, success depends on complete removal of infected and non-viable tissues and the consideration of the patient's risk factors. Unfortunately, there is no guaranteed method for assessing the accuracy of surgical debridement. Obviously, due to the operation, we are be able only to reduce the microbial load to one degree or another, but it is impossible to guarantee the removal of all dormant and sealed in the biofilm microorganisms.

CONCLUSION

Osteomyelitis remains one of the main unsolved problems in orthopedic surgery. Although significant advances have been made in recent years in understanding the pathophysiology of bone infection, there have been no fundamental changes in treatment standards. Four reservoirs of bacteria may be formed in the human body due to osteomyelitis and periprosthetic infection, the nature of each of them requires further study. In particular, it is necessary to elucidate the mechanism of *S. aureus* invasion into the lacunar-canalicular system and determine the factors leading to the ineffectiveness of the standard antibiotic therapy. Moreover, the role of intracellular persistence of bacteria in chronic osteomyelitis remains unclear due to the lack of in vivo studies, and requires further study.

REFERENCES

1. Kremers H.M., Nwojo M.E., Ransom J.E., Wood-Wentz C.M., Melton L.J. 3rd, Huddleston P.M. 3rd. Trends in the epidemiology of osteomyelitis: a population-based study, 1969 to 2009. *J. Bone Joint Surg. Am*., 2015, vol. 97, no. 10, pp. 837-845. DOI: 10.2106/JBJS.N.01350.
2. Sabah S.A., Alvand A., Price A.J. Revision knee replacement for prosthetic joint infection: Epidemiology, clinical outcomes and health-economic considerations. *Knee*, 2021, vol. 28, pp. 417-421. DOI: 10.1016/j.knee.2020.12.024.
3. Schwarz E.M., Parvizi J., Gehrke T., Aiyer A., Battenberg A., Brown S.A., Callaghan J.J., Citak M., Egol K., Garrigues G.E., Ghert M., Goswami K., Green A., Hammound S., Kates S.L., McLaren A.C., Mont M.A., Namdari S., Obremskey W.T., O'Toole R., Raikin S., Restrepo C., Ricciardi B., Saeed K., Sanchez-Sotelo J., Shohat N., Tan T., Thirukumaran C.P., Winters B. 2018 International Consensus Meeting on Musculoskeletal Infection: Research Priorities from the General Assembly Questions. *J. Orthop. Res*., 2019, vol. 37, no. 5, pp. 997-1006. DOI: 10.1002/jor.24293.
4. Hung C.H., Ko J.Y., Liao P.S., Yeh C.W., Hsu C.C., Lin M.C., Hsu H.C., Kuo S.J. Epidemiology of fatal/non-fatal suicide among patients with chronic osteomyelitis (COM): a nationwide population-based study. *J. Int. Med. Res*., 2020, vol. 48, no. 6, 300060520919238. DOI: 10.1177/0300060520919238.
5. Rimashevskii D.V., Kurmangaliev E.T., Belokobylov A.A., Batpenov N.D., Zagorodnii N.V., Malik B.K. Karagandinskii metod dvukhetapnoi revizii kolennogo sustava pri periproteznoi infektsii [The Karaganda method of two-stage revision of the knee in periprosthetic infection]. *Sbornik Nauchnykh Trudov, posviashchennyi 25-letiiu Kafedry Travmatologii i Ortopedii Rossiiskogo Universiteta Druzhby Narodov*, M., 2017, pp. 362-369. (in Russian)
6. Borsinger T.M., Pierce D.A., Hanson T.M., Werth P.M., Orem A.R., Moschetti W.E. Is the Proportion of Patients with "Successful" Outcomes After Two-stage Revision for Prosthetic Joint Infection Different When Applying the Musculoskeletal Infection Society Outcome Reporting Tool Compared with the Delphi-based Consensus Criteria? *Clin. Orthop. Relat. Res*., 2021, vol. 479, no. 7, pp. 1589-1597. DOI: 10.1097/CORR.0000000000001654.
7. Arciola C.R., An Y.H., Campoccia D., Donati M.E., Montanaro L. Etiology of implant orthopedic infections: a survey on 1027 clinical isolates. *Int. J. Artif. Organs*, 2005, vol. 28, no. 11, 1091-1100. DOI: 10.1177/039139880502801106.

8. Pulido L., Ghanem E., Joshi A., Purtill J.J., Parvizi J. Periprosthetic joint infection: the incidence, timing, and predisposing factors. *Clin. Orthop. Relat. Res*., 2008, vol. 466, no. 7, pp. 1710-1715. DOI: 10.1007/s11999-008-0209-4.
9. Kaplan S.L. Recent lessons for the management of bone and joint infections. *J. Infect*., 2014, vol. 68, no. Suppl. 1, pp. S51-S56. DOI: 10.1016/j.jinf.2013.09.014.
10. Zurawski D.V., Thompson M.G., McQueary C.N., Matalka M.N., Sahl J.W., Craft D.W., Rasko D.A. Genome sequences of four divergent multidrug-resistant Acinetobacter baumannii strains isolated from patients with sepsis or osteomyelitis. *J. Bacteriol*., 2012, vol. 194, no. 6, pp. 1619-1620. DOI: 10.1128/JB.06749-11.
11. Ramirez M.S., Bonomo R.A., Tolmasky M.E. Carbapenemases: Transforming Acinetobacter baumannii into a Yet More Dangerous Menace. *Biomolecules*, 2020, vol. 10, no. 5, pp. 720. DOI: 10.3390/biom10050720.
12. Jorge L.S., Fucuta P.S., Oliveira M.G.L., Nakazone M.A., de Matos J.A., Chueire A.G., Salles M.J.C. Outcomes and Risk Factors for Polymicrobial Posttraumatic Osteomyelitis. *J. Jt. Infect*., 2018, vol. 3, no. 1, pp. 20-26. DOI: 10.7150/jbji.22566.
13. Kluytmans J., van Belkum A., Verbrugh H. Nasal carriage of Staphylococcus aureus: epidemiology, underlying mechanisms, and associated risks. *Clin. Microbiol. Rev*., 1997, vol. 10, no. 3, pp. 505-520. DOI: 10.1128/CMR.10.3.505.
14. Otto M. Staphylococcus aureus toxins. *Curr. Opin. Microbiol*., 2014, vol. 17, pp. 32-37. DOI: 10.1016/j.mib.2013.11.004.
15. Otto M. Targeted immunotherapy for staphylococcal infections: focus on anti-MSCRAMM antibodies. *BioDrugs*, 2008, vol. 22, no. 1, pp. 27-36. DOI: 10.2165/00063030-200822010-00003.
16. Hall-Stoodley L., Costerton J.W., Stoodley P. Bacterial biofilms: from the natural environment to infectious diseases. *Nat. Rev. Microbiol*., 2004, vol. 2, no. 2, pp. 95-108. DOI: 10.1038/nrmicro821.
17. Ricciardi B.F., Muthukrishnan G., Masters E., Ninomiya M., Lee C.C., Schwarz E.M. Staphylococcus aureus evasion of host immunity in the setting of prosthetic joint infection: biofilm and beyond. *Curr. Rev. Musculoskelet. Med*., 2018, vol. 11, no. 3, pp. 389-400. DOI: 10.1007/s12178-018-9501-4.
18. Tuchscherr L., Heitmann V., Hussain M., Viemann D., Roth J., von Eiff C., Peters G., Becker K., Löffler B. Staphylococcus aureus small-colony variants are adapted phenotypes for intracellular persistence. *J. Infect. Dis*., 2010, vol. 202, no. 7, pp. 1031-1040. DOI: 10.1086/656047.
19. Pendleton J.N., Gorman S.P., Gilmore B.F. Clinical relevance of the ESKAPE pathogens. *Expert Rev. Anti Infect. Ther*., 2013, vol. 11, no. 3, pp. 297-308. DOI: 10.1586/eri.13.12.
20. Al-Maiyah M., Hemmady M.V., Shoaib A., Morgan-Jones R.L. Recurrence of chronic osteomyelitis in a regenerated fibula after 65 years. *Orthopedics*, 2007, vol. 30, no. 5, pp. 403-404. DOI: 10.3928/01477447-20070501-12.
21. Buchholz H.W., Elson R.A., Heinert K. Antibiotic-loaded acrylic cement: current concepts. Clin. Orthop. Relat. Res., 1984, no. 190, pp. 96-108.
22. Parvizi J., Gehrke T., Mont M.A., Callaghan J.J. Introduction: Proceedings of International Consensus on Orthopedic Infections. *J. Arthroplasty*, 2019, vol. 34, no. 2S, pp. S1-S2. DOI: 10.1016/j.arth.2018.09.038.
23. The role of the immune system and bone cells in acute and chronic osteomyelitis. K. Nishitani, S.N. Bello-Irizarry, K.M. Bentley, J.L. Daiss, E.M. Schwarz. In: *Osteoimmunology: Interactions of the Immune and Skeletal Systems*. 2nd Ed. J. Lorenzo, M. Horowitz, Y. Choi, H. Takayanagi, G. Schett, editors. Academic press. 2015. 376 p.
24. Kobayashi S.D., Malachowa N., DeLeo F.R. Pathogenesis of Staphylococcus aureus abscesses. *Am. J. Pathol*., 2015, vol. 185, no. 6, pp. 1518-1527. DOI: 10.1016/j.ajpath.2014.11.030.
25. Cheng A.G., DeDent A.C., Schneewind O., Missiakas D. A play in four acts: Staphylococcus aureus abscess formation. *Trends Microbiol*., 2011, vol. 19, no. 5, pp. 225-232. DOI: 10.1016/j.tim.2011.01.007.
26. Cheung G.Y., Otto M. The potential use of toxin antibodies as a strategy for controlling acute Staphylococcus aureus infections. *Expert Opin. Ther. Targets*, 2012, vol. 16, no. 6, pp. 601-612. DOI: 10.1517/14728222.2012.682573.
27. Cheng A.G., Kim H.K., Burts M.L., Krausz T., Schneewind O., Missiakas D.M. Genetic requirements for Staphylococcus aureus abscess formation and persistence in host tissues. *FASEB J*., 2009, vol. 23, no. 10, pp. 3393-3404. DOI: 10.1096/fj.09-135467.
28. Foster T.J., Geoghegan J.A., Ganesh V.K., Höök M. Adhesion, invasion and evasion: the many functions of the surface proteins of Staphylococcus aureus. *Nat. Rev. Microbiol*., 2014, vol. 12, no. 1, pp. 49-62. DOI: 10.1038/nrmicro3161.
29. Masters E.A., Trombetta R.P., de Mesy Bentley K.L., Boyce B.F., Gill A.L., Gill S.R., Nishitani K., Ishikawa M., Morita Y., Ito H., Bello-Irizarry S.N., Ninomiya M., Brodell J.D. Jr., Lee C.C., Hao S.P., Oh I., Xie C., Awad H.A., Daiss J.L., Owen J.R., Kates S.L., Schwarz E.M., Muthukrishnan G. Evolving concepts in bone infection: redefining "biofilm", "acute vs. chronic osteomyelitis", "the immune proteome" and "local antibiotic therapy". *Bone Res*., 2019, vol. 7, pp. 20. DOI: 10.1038/s41413-019-0061-z.
30. Fuqua W.C., Winans S.C., Greenberg, E.P. Quorum sensing in bacteria: the LuxR-LuxI family of cell density-responsive transcriptional regulators. *J. Bacteriol*., 1994, vol. 176, no. 2, pp. 269-275. DOI: 10.1128/jb.176.2.269-275.1994.
31. Periasamy S., Joo H.S., Duong A.C., Bach T.H., Tan V.Y., Chatterjee S.S., Cheung G.Y., Otto M. How Staphylococcus aureus biofilms develop their characteristic structure. *Proc. Natl. Acad. Sci. USA*, 2012, vol. 109, no. 4, pp. 1281-1286. DOI: 10.1073/pnas.1115006109.
32. Savage V.J., Chopra I., O'Neill A.J. Staphylococcus aureus biofilms promote horizontal transfer of antibiotic resistance. *Antimicrob. Agents Chemother*., 2013, vol. 57, no. 4, pp. 1968-1970. DOI: 10.1128/AAC.02008-12.
33. Mah T.F., O'Toole G.A. Mechanisms of biofilm resistance to antimicrobial agents. *Microbiol*., 2001, vol. 9, no. 1, pp. 34-39. DOI: 10.1016/s0966-842x(00)01913-2.
34. Putnam N.E., Fulbright L.E., Curry J.M., Ford C.A., Petronglo J.R., Hendrix A.S., Cassat J.E. MyD88 and IL-1R signaling drive antibacterial immunity and osteoclast-driven bone loss during Staphylococcus aureus osteomyelitis. *PLoS Pathog*., 2019, vol. 15, no. 4, pp. e1007744. DOI: 10.1371/journal.ppat.1007744.
35. Junka A., Szymczyk P., Ziółkowski G., Karuga-Kuzniewska E., Smutnicka D., Bil-Lula I., Bartoszewicz M., Mahabady S., Sedghizadeh P.P. Bad to the Bone: on in vitro and ex vivo microbial biofilm ability to directly destroy colonized bone surfaces without participation of host immunity or osteoclastogenesis. *PLoS One*, 2017, vol. 12, no. 1, pp. e0169565. DOI: 10.1371/journal.pone.0169565.
36. Sandbakken E.T., Witsø E., Sporsheim B., Egeberg K.W., Foss O.A., Hoang L., Bjerkan G., Løseth K., Bergh K. Highly variable effect of sonication to dislodge biofilm-embedded Staphylococcus epidermidis directly quantified by epifluorescence microscopy: an in vitro model study. *J. Orthop. Surg. Res*., 2020, vol. 15, no. 1, pp. 522. DOI: 10.1186/s13018-020-02052-3.
37. De Mesy Bentley K.L., MacDonald A., Schwarz E.M., Oh I. Chronic osteomyelitis with Staphylococcus aureus deformation in submicron canaliculi of osteocytes: a case report. *JBJS Case Connect*., 2018, vol. 8, no. 1, pp. e8. DOI: 10.2106/JBJS.CC.17.00154.
38. Masters E.A., Salminen A.T., Begolo S., Luke E.N., Barrett S.C., Overby C.T., Gill A.L., de Mesy Bentley K.L., Awad H.A., Gill S.R., Schwarz E.M., McGrath J.L. An in vitro platform for elucidating the molecular genetics of S. aureus invasion of the osteocyte lacuno-canalicular network during chronic osteomyelitis. *Nanomedicine*, 2019, vol. 21, pp. 102039. DOI: 10.1016/j.nano.2019.102039.
39. Trombetta R.P., Dunman P.M., Schwarz E.M., Kates S.L., Awad H.A. A high-throughput screening approach to repurpose FDA-approved drugs for bactericidal applications against Staphylococcus aureus small-colony variants. *mSphere*, 2018, vol. 3, no. 5, pp. e00422-18. DOI: 10.1128/mSphere.00422-18.
40. Libraty D.H., Patkar C., Torres B. Staphylococcus aureus reactivation osteomyelitis after 75 years. *N. Engl. J. Med*., 2012, vol. 366, no. 5, pp. 481-482. DOI: 10.1056/NEJMc1111493.
41. Garzoni C., Kelley W.L. Staphylococcus aureus: new evidence for intracellular persistence. *Trends Microbiol*., 2009, vol. 17, no. 2, pp. 59-65. DOI: 10.1016/j.tim.2008.11.005.
42. Clement S., Vaudaux P., Francois P., Schrenzel J., Huggler E., Kampf S., Chaponnier C., Lew D., Lacroix J.S. Evidence of an intracellular reservoir in the nasal mucosa of patients with recurrent Staphylococcus aureus rhinosinusitis. *J. Infect. Dis*., 2005, vol. 192, no. 6, pp. 1023-1028. DOI: 10.1086/432735.
43. Masters E.A., de Mesy Bentley K.L., Gill A.L., Hao S.P., Galloway C.A., Salminen A.T., Guy D.R., McGrath J.L., Awad H.A., Gill S.R., Schwarz E.M. Identification of Penicillin Binding Protein 4 (PBP4) as a critical factor for Staphylococcus aureus bone invasion during osteomyelitis in mice. *PLoS Pathog*., 2020, vol. 16, no. 10, pp. e1008988. DOI: 10.1371/journal.ppat.1008988.
44. Ellington J.K., Harris M., Hudson M.C., Vishin S., Webb L.X., Sherertz R. Intracellular Staphylococcus aureus and antibiotic resistance: implications for treatment of Staphylococcal osteomyelitis. *J. Orthop. Res*., 2006, vol. 24, no. 1, pp. 87-93. DOI: 10.1002/jor.20003.
45. Vaishya R., Agarwal A.K., Rawat S.K., Singh H., Vijay V. Is single-stage revision safe following infected total knee arthroplasty? A critical review. *Cureus*, 2017, vol. 9, no. 8, pp. e1629. DOI: 10.7759/cureus.1629.
46. Yang D., Wijenayaka A.R., Solomon L.B., Pederson S.M., Findlay D.M., Kidd S.P., Atkins G.J. Novel insights into Staphylococcus aureus deep bone infections: the involvement of osteocytes. *mBio*, 2018, vol. 9, no. 2, pp. e00415-e00418. DOI: 10.1128/mBio.00415-18.
47. Thwaites G.E., Gant V. Are bloodstream leukocytes Trojan Horses for the metastasis of Staphylococcus aureus? *Nat. Rev. Microbiol*., 2011, vol. 9, no. 3, pp. 215-222. DOI: 10.1038/nrmicro2508.
48. Zhu H., Jin H., Zhang C., Yuan T. Intestinal methicillin-resistant Staphylococcus aureus causes prosthetic infection via 'Trojan Horse' mechanism: Evidence from a rat model. *Bone Joint Res*., 2020, vol. 9, no. 4, pp. 152-161. DOI: 10.1302/2046-3758.94.BJR-2019-0205.R1.

49. Shorr A.F., Tabak Y.P., Killian A.D., Gupta V., Liu L.Z., Kollef M.H. Healthcare-associated bloodstream infection: a distinct entity? Insights from a large U.S. database. *Crit. Care Med*., 2006, vol. 34, no. 10, pp. 2588-2595. DOI: 10.1097/01.CCM.0000239121.09533.09.
50. Hamza T., Li B. Differential responses of osteoblasts and macrophages upon Staphylococcus aureus infection. *BMC Microbiol*., 2014, vol. 14, pp. 207. DOI: 10.1186/s12866-014-0207-5.
51. Holtfreter S., Nguyen T.T., Wertheim H., Steil L., Kusch H., Truong Q.P., Engelmann S., Hecker M., Völker U., van Belkum A., Bröker B.M. Human immune proteome in experimental colonization with Staphylococcus aureus. *Clin. Vaccine Immunol*., 2009, vol. 16, no. 11, pp. 1607-1614. DOI: 10.1128/CVI.00263-09.
52. Verkaik N.J., Boelens H.A., de Vogel C.P., Tavakol M., Bode L.G., Verbrugh H.A., van Belkum A., van Wamel W.J. Heterogeneity of the humoral immune response following Staphylococcus aureus bacteremia. *Eur. J. Clin. Microbiol. Infect. Dis*., 2010, vol. 29, no. 5, pp. 509-518. DOI: 10.1007/s10096-010-0888-0.
53. Muthukrishnan G., Beck C.A., Owen J.R., Xie C., Kates S.L., Daiss J.L. Serum antibodies against Staphylococcus aureus can prognose treatment success in patients with bone infections. *J. Orthop. Res*., 2020, 10.1002/jor.24955. DOI: 10.1002/jor.24955.
54. Kavanagh N., Ryan E.J., Widaa A., Sexton G., Fennell J., O'Rourke S., Cahill K.C., Kearney C.J., O'Brien F.J., Kerrigan S.W. Staphylococcal Osteomyelitis: Disease Progression, Treatment Challenges, and Future Directions. *Clin. Microbiol. Rev*., 2018, vol. 31, no. 2, pp. e00084-17. DOI: 10.1128/CMR.00084-17.
55. Expert Panel on Musculoskeletal Imaging:, Beaman F.D., von Herrmann P.F., Kransdorf M.J., Adler R.S., Amini B., Appel M., Arnold E., Bernard S.A., Greenspan B.S., Lee K.S., Tuite M.J., Walker E.A., Ward R.J., Wessell D.E., Weissman B.N. ACR Appropriateness Criteria® Suspected Osteomyelitis, Septic Arthritis, or Soft Tissue Infection (Excluding Spine and Diabetic Foot). *J. Am. Coll. Radiol*., 2017, vol. 14, no. 5S, pp. S326-S337. DOI: 10.1016/j.jacr.2017.02.008.
56. Palestro C.J., Love C., Miller T.T. Infection and musculoskeletal conditions: Imaging of musculoskeletal infections. *Best Pract. Res. Clin. Rheumatol*., 2006, vol. 20, no. 6, pp. 1197-1218. DOI: 10.1016/j.berh.2006.08.009.
57. Kapoor A., Page S., Lavalley M., Gale D.R., Felson D.T. Magnetic resonance imaging for diagnosing foot osteomyelitis: a meta-analysis. *Arch. Intern. Med*., 2007, vol. 167, no. 2, pp. 125-132. DOI: 10.1001/archinte.167.2.125.
58. Khawaja A.Z., Cassidy D.B., Al Shakarchi J., McGrogan D.G., Inston N.G., Jones R.G. Revisiting the risks of MRI with Gadolinium based contrast agents – review of literature and guidelines. *Insights Imaging*, 2015, vol. 6, no. 5, pp. 553-558. DOI: 10.1007/s13244-015-0420-2.
59. Langsjoen J., Neuwelt A., Eberhardt S., Mlady G., Shukla U., Murali S., Pizanis C., Sillerud L.O. A comparison of ferumoxytol with gadolinium as contrast agents for the diagnostic magnetic resonance imaging of osteomyelitis. *Magn. Reson. Imaging*, 2020, vol. 71, pp. 45-54. DOI:10.1016/j.mri.2020.04.012.
60. Jennin F., Bousson V., Parlier C., Jomaah N., Khanine V., Laredo J.D. Bony sequestrum: a radiologic review. *Skeletal Radiol*., 2011, vol. 40, no. 8, pp. 963-975. DOI: 10. 1007/s00256-010-0975-4.
61. Chai J.W., Hong S.H., Choi J.Y., Koh Y.H., Lee J.W., Choi J.A., Kang H.S. Radiologic diagnosis of osteoid osteoma: from simple to challenging findings. *Radiographics*, 2010, vol. 30, no. 3, pp. 737-749. DOI2: 10.1148/rg.303095120.
62. Amsilli M., Epaulard O. How is the microbial diagnosis of bacterial vertebral osteomyelitis performed? An 11-year retrospective study. *Eur. J. Clin. Microbiol. Infect. Dis*., 2020, vol. 39, no. 11, pp. 2065-2076. DOI: 10.1007/s10096-020-03929-1.
63. Bori G., McNally M.A., Athanasou N. Histopathology in Periprosthetic Joint Infection: When will the Morphomolecular Diagnosis be a Reality? *Biomed. Res. Int*., 2018, vol. 2018, pp. 1412701. DOI: 10.1155/2018/1412701.
64. Lonner J.H., Desai P., Dicesare P.E., Steiner G., Zuckerman J.D. The reliability of analysis of intraoperative frozen sections for identifying active infection during revision hip or knee arthroplasty. *J. Bone Joint Surg. Am*., 1996, vol. 78, no. 10, pp. 1553-1558. DOI: 10.2106/00004623-199610000-00014.
65. Hellebrekers P., Rentenaar R.J., McNally M.A., Hietbrink F., Houwert R.M., Leenen L.P.H., Govaert G.A.M. Getting it right first time: The importance of a structured tissue sampling protocol for diagnosing fracture-related infections. *Injury*, 2019, vol. 50, no. 10, pp. 1649-1655. DOI:10.1016/j.injury.2019.05.014.
66. Congedi S., Minotti C., Giaquinto C., Da Dalt L., Donà D. Acute infectious osteomyelitis in children: new treatment strategies for an old enemy. *World J. Pediatr*., 2020, vol. 16, no. 5, pp. 446-455. DOI: 10.1007/s12519-020-00359-z.
67. Alamanda V.K., Springer B.D. The prevention of infection: 12 modifiable risk factors. *Bone Joint J*., 2019, vol. 101-B, no. 1_Supple_A, pp. 3-9. DOI: 10.1302/0301-620X.101B1.BJJ-2018-0233.R1.
68. Sparenberg S., Blankensteijn L.L., Ibrahim A.M., Peymani A., Lin S.J. Risk factors associated with the development of sepsis after reconstructive flap surgery. *J. Plast. Surg. Hand Surg*., 2019, vol. 53, no. 6, pp. 328-334. DOI: 10.1080/2000656X.2019.1626738.
69. Kim D.Y., Kim U.J., Yu Y., Kim S.E., Kang S.J., Jun K.I., Kang C.K., Song K.H., Choe P.G., Kim E.S., Kim H.B., Jang H.C., Jung S.I., Oh M.D., Park K.H., Kim N.J. Microbial Etiology of Pyogenic Vertebral Osteomyelitis According to Patient Characteristics. *Open Forum Infect. Dis*., 2020, vol. 7, no. 6, ofaa176. DOI:10.1093/ofid/ofaa176.
70. Eades C., Hughes S., Heard K., Moore L.S.P. Antimicrobial therapies for Gram-positive infections. *Clin. Pharmacist*, September 2017, Vol 9, no 9. DOI:10.1211/PJ.2017.20203363.
71. Connelly C.M., Moon M.H., Schneekloth J.S. Jr. The Emerging Role of RNA as a Therapeutic Target for Small Molecules. *Cell Chem. Biol*., 2016, vol. 23, no. 9, pp. 1077-1090. DOI: 10.1016/j.chembiol.2016.05.021.
72. Seyler T.M., Moore C., Kim H., Ramachandran S., Agris P.F. A New Promising Anti-Infective Agent Inhibits Biofilm Growth by Targeting Simultaneously a Conserved RNA Function That Controls Multiple Genes. *Antibiotics* (Basel), 2021, vol. 10, no. 1, pp. 41. DOI: 10.3390/antibiotics10010041.
73. El-Mowafy S.A., Abd El Galil K.H., El-Messery S.M., Shaaban M.I. Aspirin is an efficient inhibitor of quorum sensing, virulence and toxins in Pseudomonas aeruginosa. *Microb. Pathog*., 2014, vol. 74, pp. 25-32. DOI: 10.1016/j.micpath.2014.07.008.
74. Jiang Y., Wang S.N., Wu H.T., Qin H.J., Ren M.L., Lin J.C., Yu B. Aspirin alleviates orthopedic implant associated infection. *Int. J. Mol. Med*., 2019, vol. 44, no. 4, pp. 1281-1288. DOI: 10.3892/ijmm.2019.4298.
75. Wei Y.P., Chien J.C., Hsiang W.H., Yang S.W., Chen C.Y. Aspirin administration might accelerate the subsidence of periprosthetic joint infection. *Sci. Rep*., 2020, vol. 10, no. 1, pp. 15967. DOI:10.1038/s41598-020-72731-y.
76. Guo P., Xue H.Y., Buttaro B.A., Tran N.T., Wong H.L. Enhanced eradication of intracellular and biofilm-residing methicillin-resistant Staphylococcus aureus (MRSA) reservoirs with hybrid nanoparticles delivering rifampicin. *Int. J. Pharm*., 2020, vol. 589, 119784. DOI: 10.1016/j.ijpharm.2020.119784.
77. Bagnoli F., Bertholet S., Grandi G. Inferring reasons for the failure of Staphylococcus aureus vaccines in clinical trials. *Front Cell Infect. Microbiol*., 2012, vol. 2, pp. 16. DOI: 10.3389/fcimb.2012.00016.
78. Akhtyamov I.F., Kuzmin I.I. *Oshibki i oslozhneniia endoprotezirovaniia tazobedrennogo sustava: ruk. dlia vrachei* [Errors and complications of the hip arthroplasty: guide for physicians]. Kazan, Tsentr Operativnoi Pechati, 2006, 328 p. (in Russian)
79. Van Vugt T.A.G., Arts J.J., Geurts J.A.P. Antibiotic-loaded Polymethylmethacrylate beads and spacers in treatment of orthopedic infections and the role of biofilm formation. *Front Microbiol*., 2019, vol. 10, pp. 1626. DOI: 10.3389/fmicb.2019.01626.

The article was submitted 24.12.2020; approved after reviewing 30.04.2021; accepted for publication 23.08.2021.

Information about the authors:

1. Denis V. Rimashevskiy – M.D., drimashe@yandex.ru, https://orcid.org/0000-0001-8122-5815;
2. Ildar F. Akhtiamov – Doctor of Medical Sciences, Ph.D., Professor, yalta60@mail.ru, https://orcid.org/0000-0001-5264-5634;
3. Pavel N. Fedulichev – M.D., p.fedulichev@gmail.com;
4. Wessam Zaalan – M.D., wsameeexx@gmai.com;
5. Kamil' A. Ustazov – M.D., ustazovkama@gmail.com;
6. Abdul Basith – M.D., braddulbasi@gmail.com;
7. Jumakhan M. Moldakulov – M.D., moldakulov67@mail.ru;
8. Maxim P. Zinoviev – M.D., max_travma@mail.ru.