V. Dubrovin, V. Gavrilyuk, O. Osadcha

WAYS TO OVERCOME THE RESISTANCE OF MICROBES TO ANTIBIOTICS

Withoud a doubt, antibiotics made a huge impact on population of people and our health. The risk of being unprotected against microbial diseases rises with each year, the overuse and incorect use of antibiotics led us here. But what exactly do microbes do to protect themeselves? There are 3 major mechanisms they use: β-lactamase inhibitors, efflux pump inhibitors (EPIs) and membrane permeabilizers.

β-lactam antibiotics inhibit proteins involved in peptidoglycan synthesis that is a part of cell wall of gram-positive and gram-negative bacteria. Penicillin and cephalosporin belong to this group of antibiotics, they are also most widely used in the world. Microbes use special enzymes against these antibiotics and were called β-lactamases. The latter divides β-lactamases into four distinct classes, termed A, B, C and D, identified on the basis of specific sequence motifs but also distinguished by fundamental differences in hydrolytic mechanism [1, p.3474]. There is another division in which classes A, C and D use serine as the reaction nucleophile and hydrolyze β-lactams via a covalent acylenzyme intermediate. Class B is in the metalloenzymes group, according to their name they use metal-activated water nucleophile to initiate hydrolytic reaction. To counter these enzymes, we use β-lactamase inhibitors. Currently available β-lactamase inhibitors only work against serine β-lactamases, whereas metallo-β-lactamases continue to spread, evolve, and confer resistance to all β-lactams, including carbapenems [2, p.1].

However, there are some means hospitals can use against classes A, C and D. Clavulanic acid has been shown to be very effective since discovery. Also, penicillin-inhibitor combinations are widely used, but they do not have that big spectrum of activity. *Klebsiella pneumoniae* carbapenemase (KPC) is microbe with class A enzymes caused a disaster in USA when it first occurred in hospital in 2011. Fortunately, the disease disappeared as quickly as it appeared, but doctors are still

worried about antibiotic resistance of microbes. Nowadays, we can treat KPC with combinations of antibiotics and inhibitors, but this therapy may do harm to a patient. New group of drugs called diazabicyclooctanones (DBOs) and avibactam was the first β-lactamase inhibitor in this group and the first great success to DBOs. Avibactam is another drug that works on A (including KPC), C and D classes. In addition, there are cases when avibactam was a weak inhibitor for class B. Avibactamis a mechanism-based non-β-lactam β-lactamaseinhibitor, based around a bicyclic core structure, that is able to acylate the active site of serine β-lactamasesin a reversible manner [1, p. 3489]. The success of DBOs has led to creating and developing alternatives of avibactam. Vaborbactam β-lactamase inhibitor contains boron and has a cyclic structure. In future we might see more boronate-based compounds inhibitors, because vaborbactam with antibiotic meropenem was effective in treatment of urinary tract infections caused by microbes with class A enzymes. They also have a potential to deal with class B for which we do not have strong inhibitors. Also, phosphonate-based compounds have been shown to inhibit some class B enzymes.

Other ways of fighting resistant microbes include strong, antibiotics like colistin, new drugs and absolutely new methods. Colistin is antibiotic that always considered with neurotoxicity and nephrotoxicity but side effects are reversible after cessation of the drug. Some scientists think colistin should become a first-line treatment for carbapenem (commonly used antibiotic) resistant gram-negative microbes such as *Enterobacterales* and *Pseudomonas aeruginosa*. It is hard to detect microbes sensitive to colistin that why antibiotic is used rarely and the second reason is earlier mentioned toxicity. The use of colistin in CR-GNB infections should be patient-specific [3, p.5]. New antibiotics are currently in clinical trials, they are designed for multidrug resistant bacterial strains, especially for *Neisseria gonorrhoeae* and *Clostridium difficile*. They include tetracycline derivatives (eravacycline), fourth generation fluoroquinolones (delafloxacin), new combinations between one β-lactam and one β-lactamase inhibitor (meropenem and vaborbactam), siderophore cephalosporins (cefiderocol), new aminoglycosides (plazomicin), and agents in development for treating drug-resistant TB (pretomanid) [4, p.1]. Without a doubt,

people should have more alternatives and not only in drugs but in approaches to treatment. Nanoparticles are considered for delivering antibiotics without activation of enzymes. In one experiment (2018) there have been constructed rhamnolipid-coated silver and iron oxide nanoparticles, they have shown to be effective against S. aureus and P. aeruginosa biofilms. Microbes evolve so fast that it's hard to be ahead of them for us, but viruses will remain the fastest to adapt. We can use bacteriophages (viruses of microbe) to target specific pathogen, this way no harm will be caused to patients. There are a lot of documented cases of phage therapy treatment. The main problem of this therapy is delivery of phages to microbes, many factors influence phages before they get to pathogen. Despite the fact that bacteriophage therapies are safe for a patient, none of them are approved by FDA. Microbes and phages are natural rivals, they both compete for existence and microbes developed their own defensive mechanism, on of them is CRISPR-Cas system. CRISPR-Cas systems are able to edit genome, they are used to create genome modified organisms (GMO). A number of CRISPR-Cas systems have been recently investigated as alternative antibiotics by reprogramming them to target bacterial DNA/RNA [5, p. 10]. In several experiments CRISPR-Cas system was delivered to multidrug-resistant bacteria and *Staphylococcus aureus* by bacteriophages and bacterial plasmids, the result was a success in both cases.

To sum up, antimicrobial resistance to antibiotics is a serious problem humanity faces right now. We already understand the mechanisms that give microbes protection from antibiotics like β-lactamase enzymes. They fall into 2 major groups serine-β-lactamases (classes A, C and D) and metallo-β-lactamases (class B). Currently we do not have anything better than avibactam to inhibit class B. However, different combinations of antibiotics and β-lactamase inhibitors are used against classes A, C and D, like clavulanic acid, penicillin-inhibitor, the same avibactam, boronate-based and phosphonate-based compounds. Other methods should be also considered in new era of antibiotics: usage of strong and slightly dangerous antibiotics (colistin), bacteriophages that are safe but hard to deliver and finally CRISPR-Cas systems to modify genome of microbes.

REFERENCES

1. Tooke L. Catherine, Hinchliffe Philip et al; β-Lactamases and β-Lactamase Inhibitors in the 21st Century; Journal of molecular biology; Volume 431, Issue 18, 23 August 2019, Pages 3472-3500
2. Mojica F. Maria, Rossi Maria-Augustina et al; The urgent need for metallo-β-lactamase inhibitors: an unattended global threat; Personal View; Volume 22, Issue 1, E28-E34, January 2022
3. Aslan, A T.; Akova, M. The Role of Colistin in the Era of New β-Lactam/β-Lactamase Inhibitor Combinations. Antibiotics 2022, 11, 277. https://doi.org/10.3390/antibiotics11020277
4. Terreni M, Taccani M, Pregnolato M. New Antibiotics for Multidrug-Resistant Bacterial Strains: Latest Research Developments and Future Perspectives. Molecules. 2021 May 2;26(9):2671. doi: 10.3390/molecules26092671.
5. Shim, H. Three Innovations of Next-Generation Antibiotics: Evolvability, Specificity, and Non-Immunogenicity. Antibiotics 2023, 12, 204. https://doi.org/10.3390/antibiotics12020204

L. Filins'kyy, O. Drobakhin, O. Hurko

EXPERIMENTAL SETUP FOR THE STUDY OF LIQUID FOAMS

Many new publications in the study of the propagation of electromagnetic waves different ranges in foam structures bring a growing interest for specialists [3]. For example, S. Kharkovsky, F. Hepburn, J. Walker and R. Zoughisolved many problems with foam coatings for modern space shuttle using millimeter waves [6], V. Alekseev, O. Drobakhin and L. Filinskyy describe the possibility of using the millimeter ranges in practical study of foam for possible applications [1].

Very important problems of the technique for measuring and calculating the dielectric properties ε and tan δ in foam plastic samples located in sections of rectangular waveguides the author reviewed in [4].

V. Neagu in his work draws the readers' attention to various issues related to foam that he solved. In particular, he pays a great attention to the equipment and methods of microwave quality control of the used foam [7].

In the microwave range of 8-12 GHz, based on the experimental data were calculated value of the reflection and attenuation of electromagnetic waves in work [5].