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# ANTIMICROBIAL PROPERTIES OF SOME PORPHYRIN COMPOUNDS AGAINST CATHETER RELATED INFECTIONS

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Summary

Central venous access is commonly used in clinical practice for a wide spectrum of procedures. The main complications limiting use of central and peripheral catheters include catheter-related infections. The epidemic of microbial resistance to widely-used antibiotic groups seriously complicates treatment of CRI

even in complex with systemic antibiotic therapy. In this paper we propose to consider porphyrin compounds as alternative antimicrobial agents for antimicrobial lock therapy. It was discovered antimicrobial action of porphyrin compounds against *S. aureus* and *E. coli* that was significantly enhanced in composition with EDTA as additional component of antimicrobial lock solution.

*Key words*: porphyrins, catheter-related infections, lock therapy, biofilm.

It is difficult to imagine modern medicine without permanent intravenous approach. There are various categories of patients whose lives depend on manipulations that require placing central or peripheral catheters. Despite of benefits use of intravenous catheters is closely related with several adverse effects. The most important one is development of the catheter-related infection (CRI) which becomes possible after catheter contamination. Two ways of catheter contamination are possible; the first one is extrinsic when catheter becomes contaminated from medical worker's hands or infusion of infected fluids. The second one is intrinsic, in this case contamination occurs with pathogens from the skin of the patient or pathogens that spreading from a distant focuses of chronic infection within patient's organism by hematogenous way. The most common pathogens that cause CRI are coagulase-negative microorganisms, *S. aureus*, C. *albicans*, *P. aeruginosa*, *E. coli* [6].

Biofilm formation occurs after microorganisms' interaction with catheter. This process is almost inevitable and depends on several factors such as material of a catheter that can promote more active adhesion of bacteria, rate of infusion and properties of infused fluids. Bacterial attachment to catheter surfaces occurs by interaction of MSCRAMMs (microbial surface components recognizing adhesive matrix molecules) with fibronectin, fibrinogen, collagen, and heparin that cover introduced catheter immediately after installation [3].

Biofilm, for its part, represent a form of adaptive resistance resulting in significant reduction of antibiotic susceptibility. It becomes possible due to biofilm's structure, especially matrix, which diminishes diffusion of antimicrobial agent through biofilm and binds with them. Persisters as part of biofilm composition also

contribute to biofilms multiresistance [1]. Thus, development of the new antimicrobial agents that could be effective in antimicrobial lock therapy composition is the question of current interest.

Antimicrobial lock therapy (ALT) – method for catheter lumen sterilizing that involves injection of high-concentrated antimicrobial agent into the catheter lumen for extended periods of time. The antimicrobial lock solution consists of two components, antimicrobial agent that often combined with an additional component, in particular anticoagulant such as heparin.

The purpose of our research is a characterization of antimicrobial properties of porphyrin compounds as potential agents for antimicrobial lock solution. The biofilm was obtained by *S. aureus* ONU 536 and *E. coli* ONU 458 that were cultivated in Peptone Glucose Water during 24 and 48 hrs at 37 °C with adding of porphyrins and EDTA in polysterene microtiter plate. The Biofilm growth intensity was tested by measuring of optical density by automatic reader «μQuant» BioTek (USA).

Porphyrins – heterocyclic compounds that take part in biochemical processes in living organisms but there are a lot of their synthetic analogous. There are two classes of porphyrins, natural (NP) and synthetic (SP), difference based on their structure and origin. Synthetic porphyrins are derived from the tetrapyridylporphyrin skeleton (SP), which has pyridyl groups at each of the four *meso*-positions and hydrogens on all the pyrrole positions [4]. The studied porphyrins (protoporphyrin IX, hemin, tetrapyridylporphyrin SP, mono-nonyl-SP, tetra-nonyl-SP, nonyl-SP-Zn) were tested in different concentrations: 0.1, 1, 10 μM.

It was discovered that regarding *S. aureus* the most effective porphyrin compound is mono-nonyl-SP at concentration 10 μM. NPs also demonstrate antimicrobial activity but less potent. As to *E. coli* cultures, mono-nonyl-SP and nonyl-SP-Zn demonstrate the most potent antimicrobial influence at concentration 10 μM. NPs don't express antimicrobial activity, they even stimulate growth of biofilm as you can see on diagram.

Antimicrobial properties of porphyrins are based on their similarity with heme. Heme is a source of iron, cofactor which takes part in electrons transferring, reactions

of peroxidase and oxidase catalysis and photons absorption [1]. In Gram-negative and Gram-positive microorganisms heme capture occurs via active transport, heme binds with TonB- ExbB- and ExbD-dependent receptor on bacterial surface. Porphyrins exploit the same path. They enter the cell and affect bacterial heme-catalysed reactions preventing their normal function in the cell metabolism. Development of resistance is very unlikely, rare bacterial mutants that become resistant to porphyrins via loss of heme-uptake systems would be less virulent. This would minimize the impact of resistance since such mutants should survive less well in body fluids and tissues. Furthermore, porphyrins can also penetrate the outer membrane independently of the heme transporters due to their hydrophobic nature and their preferential association with lipid membranes [5].

Thus, we selected two potential porphyrin compounds that we tested in composition with EDTA for the next stage of our research. We observed that antimicrobial action of porphyrins in composition with EDTA is more active compared to its separate action. Also, 48 hrs cultures become more sensitive to antimicrobial action. This synergistic effect can be explained by chelating abilities of EDTA. The affinity of EDTA toward metal ions (in particular divalent ions) determines the breakdown of a biofilm. Divalent cations such as calcium and magnesium cross-link with the polymer strands and provide greater binding force in a developed biofilm [2]. Also, EDTA has anticoagulant properties that allow substituting heparin as additional component in ALT [4].

In conclusion it should be noted, that mono-nonyl-SP and nonyl-SP-Zn was selected as the most prospective porphyrin compounds. Both demonstrate significant inhibition of biofilm growth at 24 and 48 hrs cultures. In composition with EDTA as additional compound their action was enhanced. Because of antimicrobial properties of porphyrin compounds and synergism that they demonstrate in composition with EDTA, they are promising agents for further research.

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