IMMOBILIZATION OF ANTIBACTERIAL AGENTS ON THE SURFACE OF CELLULOSE MEMBRANES MODIFIED WITH POLYGLYCIDYLMETHACRYLATE

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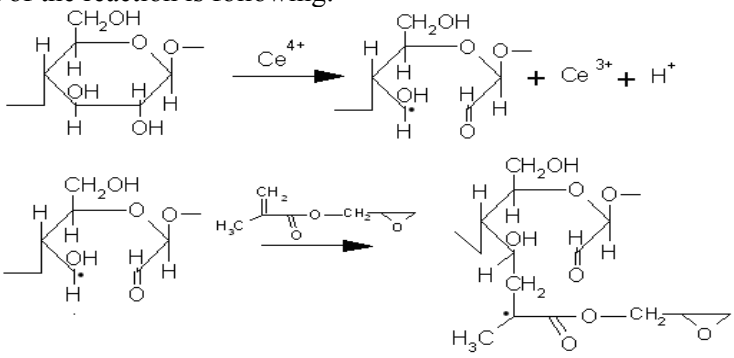
A new method of glycidylmethacrylate (GMA) radical copolymerization on the surface of commercial cellulose membranes followed by immobilization of synthetic antibiotics on grafted poly(GMA) chains has been developed. Transport and antimicrobial properties of modified membranes have been studied. It has been shown that decreasing of samples permeability is caused by grafting of GMA, and it depends on initiator concentration, duration of reaction and process temperature. Infrared spectrometry and capillary electrophoresis were used to analyze modified samples. Antibacterial properties of membranes modified by nalidixic acid and levofloxacin were studied against gram-negative bacteria E. coli and gram-positive bacteria S. aureus. Higher antibacterial activity of membranes modified by levofloxacin in comparison to those modified by nalidixic acid was shown. Thus, antibacterial activity of membranes modified by 0,5% solutions of antibiotics was 90% for samples with nalidixic acid and 100% for samples with levofloxacin. All samples showed lower antibacterial activity against gram-negative bacteria. The antibacterial activity remained stable for 36 days of membrane operation.

For the last few years the process of water treatment has become a matter of great concern for many scientists all over the world. Membrane technologies play main role among the most progressive environmentally-friendly water-purifying technologies. But there are several operational problems which limit their introduction in industrial processes. One of the main difficulties during an operation of membrane elements for water treatment is membrane biofouling [1, 2]. Typical side effects of microbiological fouling are the following: reduction of the performance of membranes due to biofilm formation on their surfaces, the secondary pollution of the purified water by bacterial cells and products of their metabolism and an increase in the power consumption because of the higher pressure requirements to overcome the biofilm resistance and the flux decline. In principle, thermal or chemical treatments are possible methods of disinfection of membrane surfaces. Thermal disinfection is not widely used and is restricted for some types of microfiltration and ultrafiltration membranes because of their low thermal resistance. On the other hand, the effective liquid disinfectants are often very aggressive, so they can cause irreversible changes to the membrane selective layer [3]. A common approach to prevent the microbiological fouling of the membranes is an appropriate preliminary treatment of feed water. This enables to decrease the content of bacteria and nutrients consumed by the bacteria during their activity in water [4]. However, such a pre-treatment is rather labour consuming and expensive. Other attempts to reduce the biofouling problem consist of specific modification of membranes aimed at alteration of the membrane surface chemistry [5–10].

In this paper a new way of biocide substance introducing on a membrane surface is proposed. The goal of this work was to produce membranes resistant to biofouling by grafting of glycidylmethacrylate (GMA) with the next immobilization of antibacterial agents on grafted chains. The molecule of monomer GMA contains two functional groups: double bond and epoxy group. Due to its bifunctionality GMA was chosen to modify cellulose membranes. On the one hand, double bond enables process of polymerization and grafting of GMA, on the other hand, epoxy group can form covalent bonds with different substances, among which are carbon acids, amines, halides and others [10]. These properties of GMA were used in our work.

Manufactured asymmetric cellulose membranes were modified with GMA using method of radical graft copolymerization. Reaction was performed in aqueous medium. Cerium ammonium nitrate was used as initiator. The chemism of the reaction is following:



The efficiency of GMA grafting on the membrane surface was evaluated indirectly with the help of water-flux measurement method. It is known [12] that grafting of polymer chains results in decreasing of effective pore size. This process causes the decline in water flux trough modified membranes. Therefore the effectiveness of GMA grafting was evaluated by measuring the changes in water flux values (productivity,

Jv) and comparing them to those for unmodified samples. The process of monomer reaction with cellulose surface depends on different factors. The main roles play concentration of initiator, temperature, and duration of modification process. Series of experiments allowed do determine optimum reaction conditions and to prove that temperature variation in 25 degrees does not have considerable influence on grafting effectiveness.

Infrared spectra of modified cellulose membranes were made in order to confirm qualitatively presence of polyGMA chains. Appearance of new absorption bands at 1745 cm^{-1}, 1255 cm^{-1} and 925 cm^{-1} in IR-spectrum of modified sample correspond to stretching vibrations of C=O and epoxy-groups in grafted polyGMA; Thus, the results of IR-spectroscopy examination proved the presence of polyGMA chains in modified samples.

The samples of membranes with poly(GMA) chains grafted onto their surfaces were used to immobilize synthetic antibiotics nalidixic acid and levofloxacin. The process of immobilization was based on covalent binding of carboxylic groups of antibacterial agents with epoxy groups of poly(GMA). The concentration of antibiotics varied from 0,01 to 1%. After antibiotics immobilization adsorption/desorption of nalidixic acid and levofloxacin on cellulose membranes were examined using the system of capillary electrophoresis. The data obtained from electron electrophoresis show that the amount of immobilized antibiotics at 1 cm^2 of membrane surface depends on concentration of its solutions. Desorption of immobilized antibacterial agents in aqueous medium was absent during 28 days of observations. Antibacterial activity investigation of modified samples was held using the method which was reported earlier [11]. The strains, taken from Ukrainian Collection of Microorganisms, *Escherichia coli* BE, *Escherichia coli* HB 101, *Staphylococcus aureus CCM 485* were used in order to control antibacterial activity of membranes. The results show that for both antibacterial agents it is lower against gram-positive bacteria. Thus, samples modified with 1% levofloxacin solution displayed 100% antibacterial activity against *E. coli* (Fig. 1a) and 88% activity against *S. aureus* (Fig. 1b).

Antibacterial activity investigations of cellulose membranes with nalidixic acid and levofloxacin immobilized on their surface were held against *E. coli* and *S. aureus.* The results show that in both cases it is lower against gram-positive bacteria. Thus, samples modified with 1% levofloxacin solution displayed 100% antibacterial activity against *E. coli* (Fig. 1a) and 88% activity against *S. aureus* (Fig. 1b).

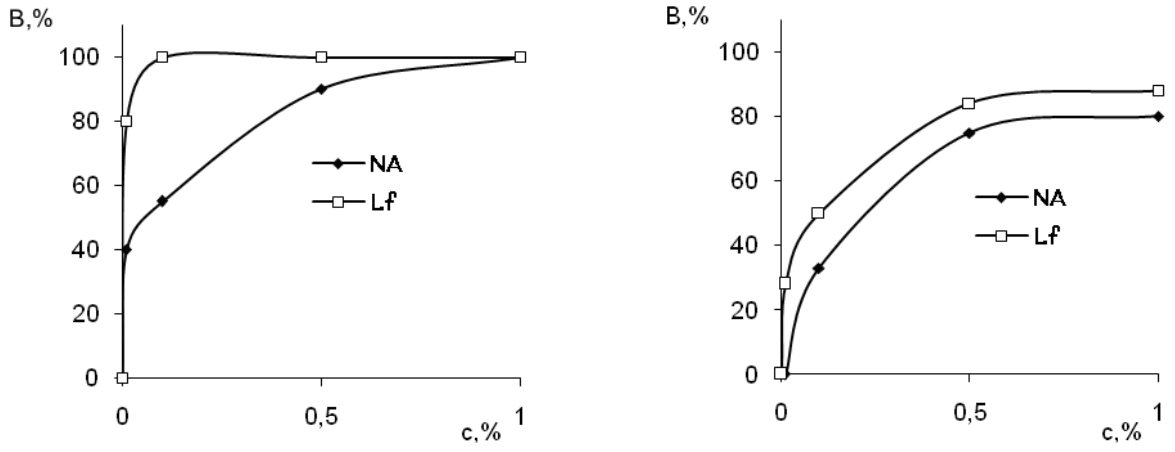


Figure1. Dependence of antibacterial activity (*B*) of cellulose membranes on nalidixic acid and levofloxacin concentrations against *E. coli (a)* and *S. aureus (b).*

As it can be seen from Fig.3, antibacterial activity of membrane samples increases when the concentration of modifying solutions and amount of immobilized antibiotics rises. Antibacterial properties of immobilized levofloxacin against both bacteria strains are expressed stronger than those of nalidixic acid, which also can be seen from Fig. 3 *a* and *b.* For instance, antibacterial activity of membrane samples after modification in 0,1 and 0,5% solutions of antibiotics is about 50 and 90% accordingly for nalidixic acid, and 99,99 and 100% for levofloxacin. Further investigations revealed that antibacterial activity of modified samples remains stable after 36 days of operation.

References

[1] Flemming H. Experimental Thermal Fluid Science ***14 (4),*** *387 (1997). [2] Vrouwenvelder H, Paassen J. Desalin.* ***118,*** *162 (1998). [3] Block S. Disinfection, Sterilization and Preservation. Malvern, PA, 1991. [4] Baker J. Desalination. 118, 81 (1998). [5] Pieracci J., Crivello J., Belfort G. J. Membr. Sci.* ***202****, 7 (2002). [6] Bryk M. Naukovi Zapysky NaUKMA.* ***18****, 4 (2000). [7] Hrasch P., Gorenflo A. Desalin.* ***172****, 41 (2005). [8] Park N., Kwon B. J. Membr. Sci.* ***258****, 43 (2005). [9] Tatsuo T. Macromol. Mat. Eng.* ***286****, 63 (2001). [10] Surovcev M., Mihlin V. VII International Conference of Oligomer Chemistry and Physical Chemistry. Moskov, (2000), 109. [11] Konovalova V., Pobigay A. Critic. Techn. Membranes* ***4****, 56 (2006). [12] Xu Z., Wang J., Shen L. J. Polym. Sci.* ***196****, 221 (2002).*