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### TILORONE EFFECT ON SALMONELLA ENTERITIDIS BIOFILM FORMATION

### Abstract

The study of how tilorone in different concentrations effect on biofilm formation ability of collection strains of Salmonella enteritidis, was performed. For the experiment were prepared five dilutions of tilorone 25, 50, 100, 250 and 500 mcg/ml. To determine the effect of tilorone on the biofilm formation used method for cultivation of the biofilms in the plates for cell culture, using the medium LB. The presence of the biological activity of the compounds judged by the presence of optical density difference between the experimental and control samples. The results showed that the different concentrations of test compound act differently on growth and biofilm formation of test strains *S. enteritidis*, causing stimulation or inhibition, or nothing changes at all.

#### Introduction

Today, epidemics of many diseases are less common now; infections caused by conditionally pathogenic microorganisms are becoming more widespread.

In the twentieth century, antimicrobial agents were created which almost immediately entered clinical practice and produced high results.

However, through the widespread use of antibiotics, science and medicine have faced a rather complex problem of resistance to various antimicrobial drugs.

Particular attention should be paid to conditionally pathogenic bacteria that very quickly acquire polyresistance to a wide range of antimicrobial agents; in particular, S. enteritidis is a microorganism that very often causes acute intestinal infections. An additional factor of danger is that it is very common bacteria, which very often present in raw food. Thus, the development and introduction of the medical practice of new approaches for combating opportunistic pathogenic microorganisms are relevant.



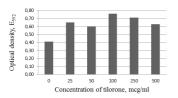
#### Materials and methods

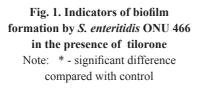
The experiment was conducted on a several strains of *Salmonella enteritidis* ONU 266, ONU 465 and ONU466.

In work used the method of culturing microorganisms in static conditions using sterile 96-well polystyrene flat-bottomed plates. To determine the intensity of biofilm formation was used the method of staining by crystal violet with some modifications. Formation of biofilm determine spectrophotometrically [1].

### **Results and discussions**

The influence of tilorone on the formation of biofilms by *S. enteritidis* ONU 266, *S. enteritidis* ONU 465 and *S. enteritidis* ONU 466 was performed after 24 hours of incubation with different concentrations of test compound. Comparison of control culture and culture with tilorone showed that stimulation of biofilm formation at all concentrations was observed only in the strain *S. enteritidis* ONU 466 and maximum gain was 85, 4%, compared with the control, at concentration of tilorone 100 mcg/ml (Figure 1).





Mainly observed stimulation effect of compound strain S. enteritidis ONU at 465 Different (Figure 2). concentrations of test compound affected differently. inhibiting and stimulating the formation of biofilm only at the strain of S. enteritidis ONU 266, but this strain was shown highest inhibition effect, about 57, 47%, compared with the control, at concentration of tilorone 50 mcg/ml (Figure 3).

The results showed that the different concentrations of test compound act differently on growth and biofilm formation of test strains *Salmonella enteritidis*, causing stimulation or inhibition, or any changes at all. But, at certain concentrations in specific strains experienced a pronounced stimulatory and/or inhibition effect on the intensity growth and formation of biofilm of microorganism. In this regard *S. enteritidis* ONU 266 was chosen as the model of inhibition effect of tilorone on ability to form biofilm, and strain *S. enteritidis* ONU 466 as model of stimulation.

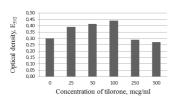


Fig. 2. Indicators of biofilm formation by *S. enteritidis* ONU 465 in the presence of tilorone Note: \* - significant difference compared with control

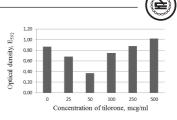


Fig.3. Indicators of biofilm formation by*S. enteritidis* ONU 266 in the presence of tilorone Note: \* - significant difference compared with control

## Conclusions

After comparison with other synthetic low-molecular inducers of interferon, for example, a derivative of acridine acetic acid, which have a pronounced antibacterial activity and suppress a number of opportunistic microorganisms in vitro, we can speak of the effectiveness of tilorone as an antibacterial drug; however, this efficiency depends on its concentration and microorganism characteristics. The conducted study of the antimicrobial properties of this compound may be the foundation for further study of its mechanisms of action on a bacterial cell or influence on the processes associated with the formation of biofilms.

### References

1. Christensen G. D. Adherence of coagulase-negative Staphylococci to plastic tissue culture plates: a quantitative model for the adherence of Staphylococci to medical devices // J. clin. microbiol. – 1985. –V. 22. –  $N_{2}$  6. – P. 996-1006.