ABSTRACT

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Title of thesis: Optimization of the method for sensitivity evaluation of biofilm-forming staphylococci against candidate antimicrobial compounds

Background: The aim of this thesis was to optimize approach for *in vitro* formation of staphylococcal biofilms on the pegs and on the wells of the 96-well panel as an analogous approach to commercially available Calgary Biofilm Device system. The aim of the Experiment 1 was to evaluate incubation conditions (such as impact of a growth medium, incubation mode, optical density of the starting bacterial inoculum and type of surface) leading to maximal biofilm formation of two biofilm producer strains with unknown biofilm phenotype and one staphylococcal strain known as strong biofilm producer. The most advisable conditions were used in incubation process of Experiment 2. This work should propose the approach leading to *in vitro* formation of the most voluminous staphylococcal biofilms exploitable for candidate drug antimicrobial activity testing.

Methods: Spectrophotometric measurement of the crystal violet colour extracted from wells with fixed and stained *Staphylococci* to evaluate the ability to form biofilm.

Main findings: The ability of three staphylococcal strains to form biofilm was affirmed**.** Soyabean medium supplemented with pig plasma, incubation with shaking and starting bacterial inoculum with optical density of 0,1 were evaluated as the most convenient conditions. In Experiment 2 staphylococcal clinical isolates were categorised according to the ability to form biofilm in conditions chosen in Experiment 1. 10 out of 25 isolates were recognised to be “Moderate biofilm producers” or “Strong biofilm producers”.

Conclusions: Using verified biofilm producers, it is possible to find the most convenient conditions for biofilm formation. Certain staphylococcal clinical isolates are capable to form biofilm voluminous enough for valid antimicrobial candidate compounds testing These isolates were categorised as moderate or strong biofilm producers.

Key words: Calgary Biofilm Device, anti-biofilm activity evaluation, biofilm-forming microorganisms, bacterial resistance vs. biofilm