

ABSTRACT

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Title: Study of the development of microbial biofilm within an experimental model, *Galleria mellonella*

Aim of the thesis: The aim of this thesis is to introduce the invertebrate organism, *Galleria mellonella* (*G. mellonella*), that can be used within the scientific research as an alternative model organism to vertebrates. This model seems to be more financially favorable and more ethically acceptable, which is an essential fact in the current era of increasing pressure to abandon testing on vertebrates in particular. The experimental part of this thesis is focused on optimizing the utilization of the *G. mellonella* larvae as a model organism within the research of fungal burn wounds fungal infections induced by the yeast *Candida albicans*. Within these experiments, larvae were used to preparation of tissue explants on which the formation of a single-species yeast biofilm was investigated.

Methods: Before obtaining of tissue explants, the larvae were decapitated. To reduce contamination of tissue samples primarily by the gut microbiota of the larvae, the approach of removing the digestive tube was optimized. Burn wounds were induced *post mortem* using a iron nail. After the burn wound infection and incubation, microscopic methods were employed for monitoring of yeast biofilm formation. Cultivation, microscopic and biochemical methods were chosen to approximate determination of microbial contamination of the wound by larval microbiota.

Results: Optimization of the methodological approach resulted in obtaining up to three representative cuticle explants from a single larva. In our experiments, we did not registered the formation of a mono-species yeast biofilm in the burn wound of a larval cuticle explant. We were able to demonstrate that after infection with the yeast *C. albicans*, the fungal agent invades into deeper layers of the tissue explant (invasive

candidiasis). We observed the presence of larval microbiota in the hemolymph as well as in the homogenate of the tissue explant.

Conclusion: According to our findings, the use of tissue explants (larval cuticle explants) for the formation of mono-species yeast biofilm communities in induced burn wounds doesn't appear to be optimal choice. It should be taken into account, that larvae of *G. mellonella* are associated with microbial load within the whole body. Based on this fact, employment of *G. mellonella* for formation of mono-species biofilm communities of selected pathogens *in vivo* is experimentally difficult.

Keywords: Biofilm, *Galleria mellonella*, *Candida albicans*