



Title: OVOBIOM: Probiotics and prebiotics, effect on the chicken intestine, health and organism performance

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Introduction: Modulation of chicken intestinal microbiota by probiotics and prebiotics, is a sustainable strategy to enhance the gut health and immunity of the chicken and a solution to combat the stresses related with intensive poultry production. The gap and lack of knowledge about proper substance for modulating the gut microbiome and tracking their function is the aim of this research to elucidating the mode of probiotic and prebiotic action in chicken gut through *in ovo* injection.

1. Which probiotics & prebiotics could be the best candidate for modulating chicken microbiome?

Pre-selection of bioactive candidate compounds (prebiotics, probiotics or synbiotics) with the highest potential for the chickens (the animal model of this project) with the help of Microplate Reader as 2 step selection.

Results: Find the promise candidates, a pre-selection in the lab to measure the kinetics of prebiotics with probiotics bacteria.



*Probiotic bacteria colony in prebiotic culture

*ADULT CHICKEN

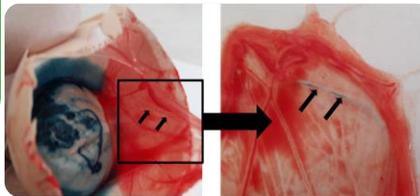


*42 DAYS CHICKEN

4. How to characterize the probiotics and their function in the chicken intestine?

Using two *in vitro* modules, Microbiological and cellular such as, antibiotic disc diffusion assay, minimum Inhibitory Concentration and antagonistic activity of probiotics.

Results: will provide detailed functional profiles of the probiotics to screen probiotics for antibiotic susceptibility and a quantitative measurement of susceptibility of probiotics to antimicrobials also, probiotics will be tested against selected pathogen strains to see the effects.



*Model of *in ovo* delivery and penetration of the bioactive solution through the chorioallantois membrane into the circulatory system of the chicken embryo (Siwek, M., et al. "Prebiotics and synbiotics-*in ovo* delivery for improved lifespan condition in chicken, 2018)

2. What dosage of our probiotic, prebiotic or synbiotics are suitable for *in ovo* injection?

The material will be fertilized eggs (n=9000) from a broiler Ross, *in ovo* injection the bioactive compound in 3 different dosage in pre hatched time (around -7+14) and pigment model of injection of prebiotics. To confirm that the inoculated bacteria are present in the hatched birds from the meconium using RT-qPCR will be performed. The QIAamp DNA Stool Minikit (Qiagen) will be used to extract DNA from meconium samples.

Results: Indicate microorganisms which will be specific for the *in ovo* stimulated microbiome and characterize the biodiversity of microbiota in the peri-hatch period and in the matured gut of the chicken.



*Manual injection of prebiotics at Day 12, UTP Archive

3. Which *in vivo* analysis will show the effect of probiotics and prebiotics on chicken microbiome and their phenotype?

Metagenomic analysis from the gut content in different time points, pre-hatch, around hatchery and at the end of the chicken life, will be based on Next Generation Sequencing (NGS) using the 16S rRNA Illumina MiSeq platform. Also, phenotypic analysis from chicken gut and common broiler pathogens.

Results: To investigate whether and how the early stimulation of a microbiome will influence chicken resistance to environmental pathogens which influence the health of broilers but also the meat for consumers.

5. Conclusion:

This study will give us a detailed functional profile of the probiotics and prebiotics and how they modulated the chicken's gut microbiome. The results will enable explain the interaction of probiotics with the cells lining the gut and with modulation the *in ovo* microbiome during peri-hatch period we will reach and promote a healthy phenotype for having the maximum beneficial function. (The research is financed under National Science Centre UMO-2019/35/B/NZ9/03186)