Intramammary Inhibition of Major Mastitis Pathogens by Non-*aureus* Staphylococci, a Mouse Model

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Mastitis is the most common and costly disease that affects dairy herds. Non-*aureus* staphylococci (NAS) are the most prevalent group of bacteria found in bovine milk; however, it is not clear whether they cause mastitis or if they confer protection against udder infection by major pathogens. Because NAS are a heterogeneous group, it is possible that their role in udder health differs among the different species and genotypes. A previous study by our research group revealed that some strains of NAS were able to inhibit the growth of *S. aureus* in vitro. Thus, we hypothesize that those NAS strains are also able to inhibit intramammary infection caused by *S. aureus*. One of the main goals of this study is to investigate if results obtained *in vitro*, which demonstrated that certain genetically similar NAS strains differ in their abilities to inhibit major pathogens, also apply *in vivo*.

To test our hypothesis, a mouse model of mastitis will be used. Three NAS strains with the greatest inhibition ability will be selected for this study. Based on the analysis of phylogenetic trees, three genetically similar NAS strains that were not able to inhibit *S. aureus* in vitro will also be selected. The ideal infection dose for each bacterial strain will be determined based on persistence and local inflammatory responses in the mammary gland, which will be assessed in a separated study. Lactating mice will be infected 14 days after giving birth in the 4^{th} mammary gland pair with 50µL of a NAS suspension followed by *S. aureus* 24h later. The mammary glands will be harvested 24 h after *S. aureus* inoculation, and the tissue homogenate will be plated in serial dilutions to assess bacterial growth. A media selective for staphylococci and differential for *S. aureus* will be used in order to differentiate between the two species infected.

This study will help to characterize the role of NAS in udder health and also represent a novel opportunity to prevent mastitis and decrease antimicrobial use, which is of great importance for the dairy industry.