## Inhibition of intramammary infection with *Staphylococcus aureus* by non-*aureus* staphylococci

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Mastitis is the most common disease in dairy herds and causes substantial economic losses to the dairy industry. *Staphylococcus aureus* is a major pathogen that causes severe and persistent udder infections that requires long therapy. Certain non-*aureus* staphylococci (NAS) strains isolated from milk samples are able to inhibit in vitro growth of *S. aureus* by means of antimicrobial peptides called bacteriocins. Thus, we hypothesize that these NAS strains are also able to inhibit udder infection caused by *S. aureus*.

To test our hypothesis, we are developing a mouse mastitis model for 3 specific NAS (*S. simulans* 3061, *S. capitis* 4231, and *S. epidermidis* 1778) and *S. aureus* in C57BL/6 mice. First, for each of the three inhibiting NAS strains, mice will receive different dose  $(1 \times 10^2, 4 \times 10^2)$ , or  $16 \times 10^2$  bacterial cells) to determine whether there will be differences in bacterial growth and inflammation among the NAS strains, and to choose the lowest dose that allows sufficient bacterial growth while causing the least inflammatory response. Then, the ideal inoculation dose for each bacterial strain will be used to conduct superinfection with *S. aureus* in the lactating mice, whereby *S. aureus* will be inoculated 24 h after inoculation with NAS.

Our results demonstrated that there is no difference of the distribution of S. simulans 3061 in mammary tissue recovered 24 h after inoculation of  $1 \times 10^2$ ,  $4 \times 10^2$ , or  $16 \times 10^2$  bacterial cells. However, inoculation of  $4 \times 10^2$  bacterial cells to mammary glands showed the most consistent results.

To evaluate the ability of certain NAS strains to inhibit udder infection caused by *S. aureus*, clinical signs, histological analysis, and bacterial load per gram of tissue will be measured. Additionally, mouse and bacterial RNA will be extracted to analyze gene and protein expression of cathelicidins and bacteriocins. Pro-inflammatory cytokines, such as IL-6, IL-1 $\beta$  a TNF- $\alpha$  will be quantified.

Implications: The possible inhibitory effects of certain NAS strains on udder infection with *S. aureus* represent an opportunity to find alternatives for the prevention and treatment of mastitis. This is of great importance not only for improving milk production and milk quality but also for the efforts to decrease the usage of antibiotics in livestock.