

Early Host-Pathogen Interactions during Mastitis in Experimentally-infected Sheep.

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Main aspects of the project

1. Develop an animal model to study mechanisms underlying resistance to mastitis
 - * Production of sheep with high and low resistance to mastitis using a divergent selection based on breeding values for SCC
2. Investigation of the inflammatory and immune responses during *Staphylococcus-induced* mastitis
 - *At the cellular levels : number and phenotypic characterization of effector cells
 - *At the biochemical level : identification of proteins that correlate with protection

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Evaluation of mastitis resistance in the high and low SCC lines

- Similar milk production but strong divergence in SCC between lines (Fig1)
- Less clinical mastitis in the Low SCC line than in the High SCC line
- Less intra mammary infections in the Low SCC line than in the High SCC line (Fig2)

Sampling time	High SCC line	Low SCC line
Backling period	~3.5	~2.5
M3	~3.5	~2.5
M4	~3.5	~2.5
M5	~4.5	~2.5
M6	~4.5	~2.5
M7	~5.5	~2.5
M8	~6.5	~3.5
M9	~4.5	~2.5

Line	Percentage
High SCC line	~45%
Low SCC line	~20%

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Experimental model of infection

- Animals: dairy ewes of the divergent lines (n=12)
 - reared at the INRA experimental farm (La Fage, France)
 - 1st lactation (highly seasonal lambing: December to February)
 - issued from a divergent selection based on SCC
- Pathogen: *Staphylococcus aureus* isolated from a chronically infected mammary gland of a ewe (leucotoxin)
- Experimental challenge with 10⁹ CFU/udder half inoculated directly into the gland cisterna

⇒ Mastitis is more severe in High SCC group

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Gene expression in milk SCC

1. Collection of Somatic Cells = neutrophils, viability>98%, isolated by centrifugation
2. Flow cytometry characterization (lymphocytes, monocytes, and granulocytes)
3. RNA extraction and quality control (Agilent bioanalyzer)
4. Hybridisation on a 20k bovine cDNA microarray (ARKs-genomics) and data analysis to be done

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