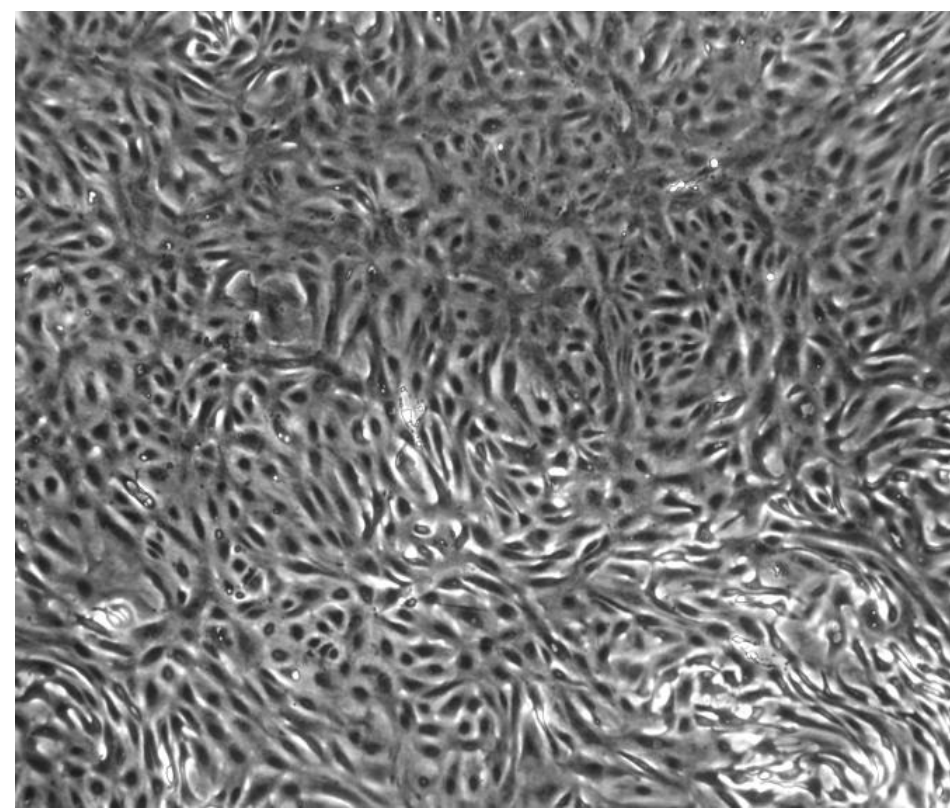


Marker selection for udder health alters the cytokine secretion of bovine mammary epithelial cells after bacterial infection

H. Kliem, M. Grandek, B. Griesbeck-Zilch, C. Kühn*, H.H.D. Meyer
Physiology Weihenstephan, Technische Universität München, Germany
*FBN, Dummerstorf, Germany

Introduction



Confluent bovine primary mammary epithelial cells

A new strategy to improve udder health is the selection of less susceptible cows using genetic markers for quantitative trait loci (QTL) on chromosome 18, which seems to influence the somatic cell count (SCS) in milk.

We established a cell culture model with bovine primary mammary epithelial cells from 28 Holstein-Friesian cows either low (Q) or high (q) susceptible to mastitis and either marker (MAS) or conventionally (CON) selected. The cells were infected for 1, 6, 24 hours (h) with heat inactivated *Staphylococcus aureus* (*S. aureus*) and *Escherichia coli* (*E. coli*) to investigate the secretion of Interleukin-8 (IL-8) as part of the innate immune system in the mammary gland.

Material and method

Starting from a large pool of heifers, 2 different selection strategies for high and low susceptibility to mastitis as reflected in SCS were applied. The conventional selection strategy relied exclusively on relative EBV for SCS (RZS) estimated in the routine breeding evaluation. In contrast to the conventional strategy, the marker assisted selection strategy primarily relied on marker information regarding paternally inherited marker alleles on chromosome 18. (Griesbeck-Zilch et al. 2009).

A human CXCL / IL-8 Elisa Kit (R&D Systems, Minneapolis, USA) was used to evaluate the protein concentration of IL-8 in the cell culture supernatants. A recombinant bovine IL-8 (Biomol, Hamburg, Germany) was used as control. The cell supernatants were lyophilised before measurement in order to obtain a higher concentration of IL-8 within the detection limit of the ELISA.

IL-8 after *E. coli* and *S. aureus* infection

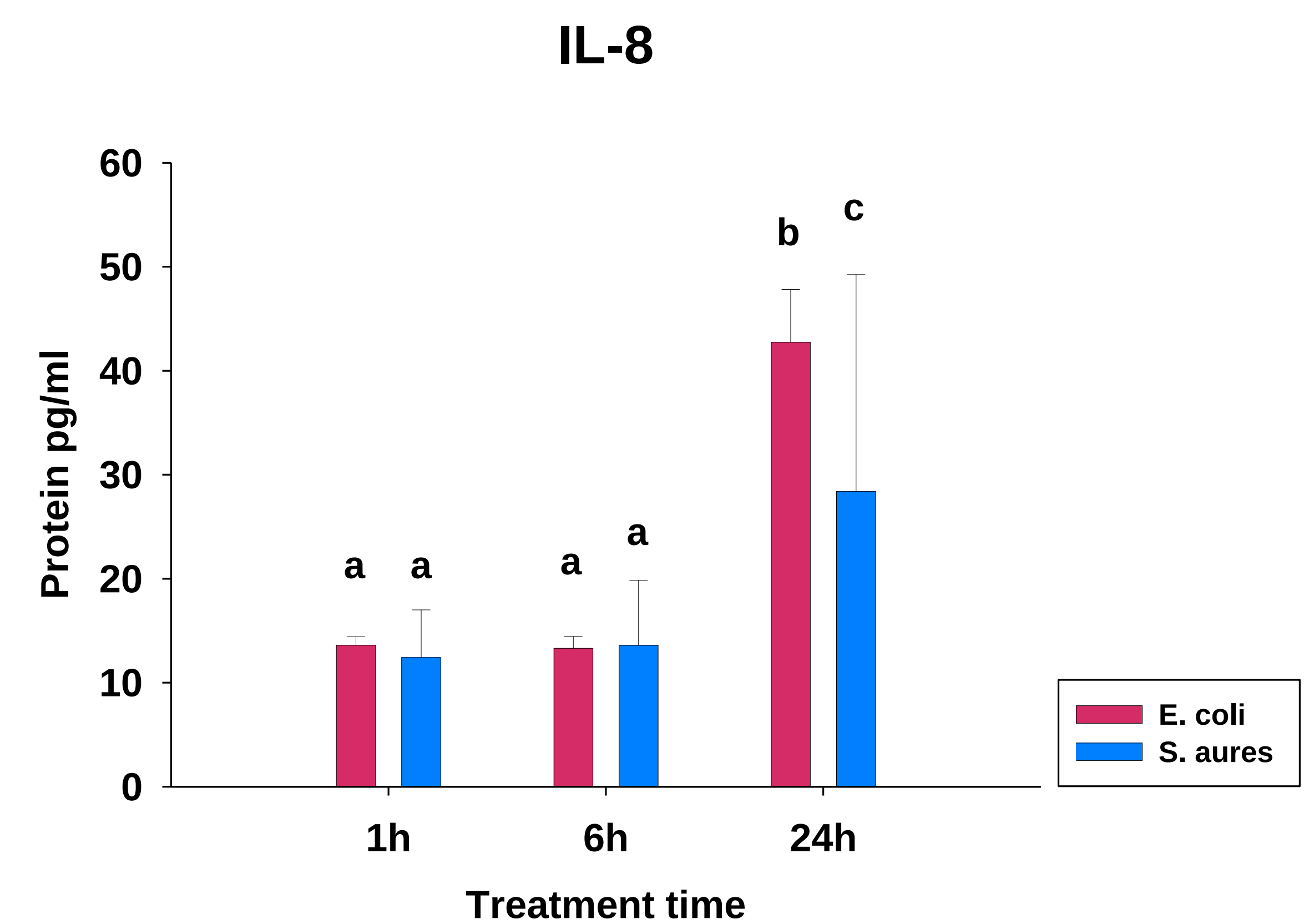


Fig. 1: Comparison of IL-8 protein concentration in mammary epithelial cells after *E. coli* and *S. aureus* treatment. Data are shown after subtraction of the control values as mean $\pm$ SEM. Different superscript letters indicate significant differences ($P < 0.05$).

Comparison of marker assisted (MAS) and conventionally (CON) selected cows

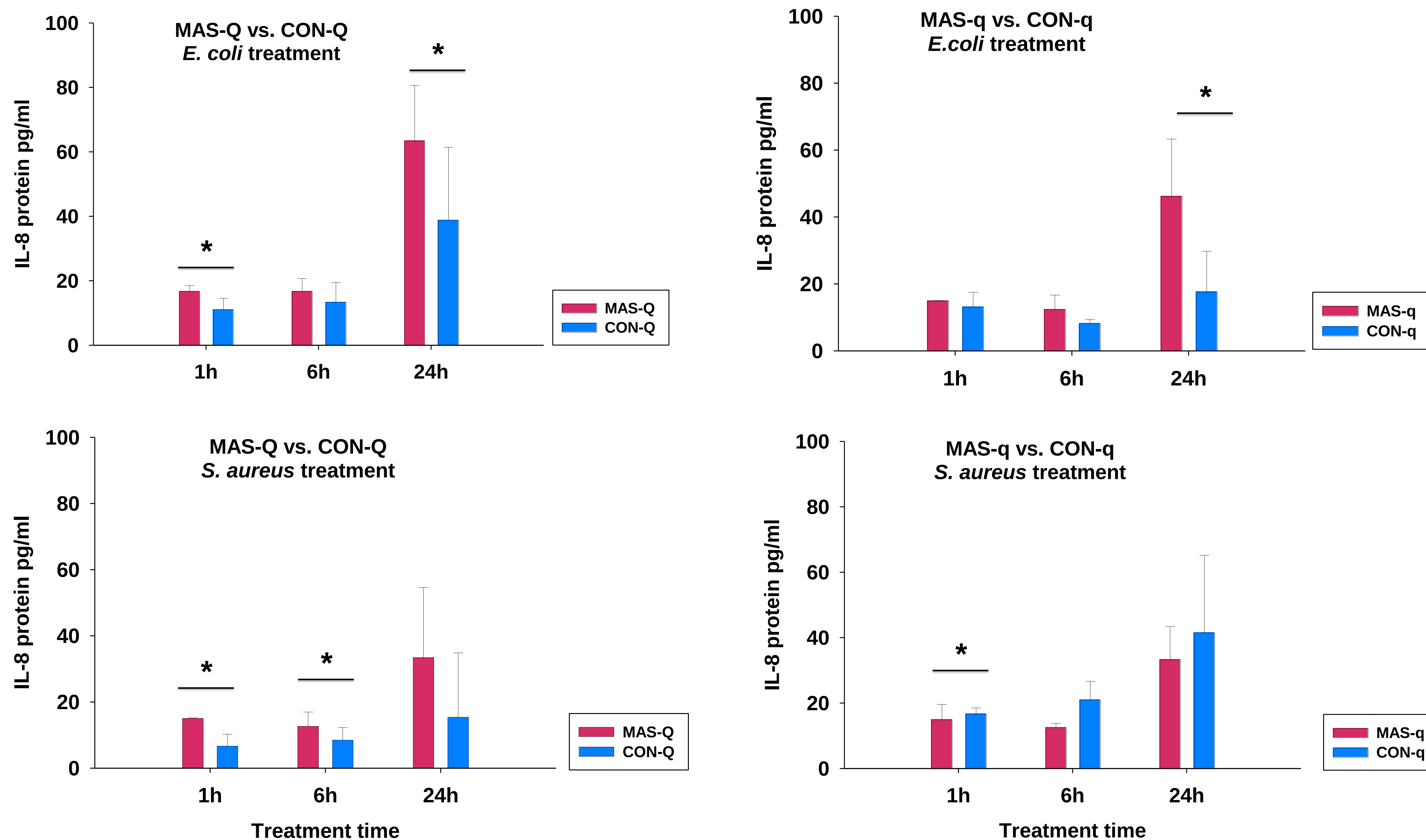


Fig. 2: Comparison of IL-8 protein concentration (after *E. coli* and *S. aureus* infection) between marker assisted (MAS) and conventionally (CON) selected cows, as well as between highly (q) and lowly (Q) susceptible cows within these two groups. Data are shown after subtraction of the control values as mean $\pm$ SEM. Significant differences within one time point are indicated by * ($P < 0.05$).

Results

A comparison between *E. coli* and *S. aureus* treatment revealed a significant increase of IL-8 after 24h for both bacteria. Furthermore the concentration of IL-8 was significantly higher after the infection with *E. coli* compared to *S. aureus* (fig.1).

A further comparison was conducted between high (q) or low (Q) susceptible cows for marker assisted (MAS) and the conventionally (CON) selected group. The results after *E. coli* treatment demonstrate a significant higher protein concentration of IL-8 in the MAS-Q compared to the CON-Q group after 1h and 24h. A similar significant increase was also detected in the MAS-q compared to the CON-q group after 24h. The protein concentrations of IL-8 after *S. aureus* infection was also higher in the MAS-Q compared to the CON-Q group after 1h and 6h. In the MAS-q vs. CON-q group only a significant difference was seen after 1h (fig. 2).

Conclusion

Breeding cows low susceptible for mastitis after marker assisted selection (MAS) seems to be a promising method against mastitis, as these animals secrete more IL-8 after a bacterial infection leading to a higher immune response than conventionally selected cows. Perhaps this is the right step towards enhanced mastitis resistance to affect animal welfare positively and lessen the need for antibiotics to treat this disease.