Antigenicity differences between Japanese and European *Staphylococcus aureus* and *Escherichia coli* bovine mastitis isolates.

Prenafeta, A.; Cesio, M.

HIPRA, Amer (Girona), Spain.

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INTRODUCTION

STARTVAC® (HIPRA) is a polyvalent mastitis vaccine, directed against both enterobacterial and staphylococcal species, that has been registered for use in the European Union and in many other countries worldwide. The STARTVAC® formulation is based in the inactivated *Escherichia coli* J5 strain and a *Staphylococcus aureus* bacterin containing the exopolysaccharide Poly-N-acetyl β-1,6 glucosamine (PNAG) involved in the biofilm phenotype of the bacteria [1]. **The objective** of this study was to analyse the reactivity of the antibodies induced by the STARTVAC® vaccine against *S. aureus* and *E. coli* bovine mastitis isolates from different areas of Japan and Europe.

MATERIALS AND METHODS

The antigenicity of *S. aureus* Japanese and European isolates was evaluated by western blot using serum from a cow vaccinated with STARTVAC®. With this purpose, cell wall-associated proteins from *S. aureus* were extracted with mutanolysin [2], analysed in SDS-PAGE and immunodetected with the specific bovine antiserum. In order to demonstrate that the antigenicity of the core lipopolysaccharide is constant in different isolates of *E. coli*, the reactivity of a specific monoclonal with several isolates of this bacterium was assessed. This was performed by exposing the monoclonal "*E. coli* J5 LPS PIERCE" to an indirect ELISA coated with different isolates of *E. coli*. In the same way, the ability of a serum from a cow vaccinated with STARTVAC® to react against several *E. coli* isolates from different origins was studied also by an indirect ELISA.

RESULTS

The *S. aureus* cell wall-associated proteins analysed by western blot showed a similar pattern between the Japanese and European isolates when the proteins were probed against antibodies induced by the STARTVAC® vaccination (Fig. 1). Concerning *E. coli* , all the Japanese and European isolates analysed by ELISA shared the same epitope in the lipid A of LPS recognised by a specific monoclonal antibody and, moreover, antibodies induced by STARTVAC® vaccination reacted against all the isolates (Fig. 2).

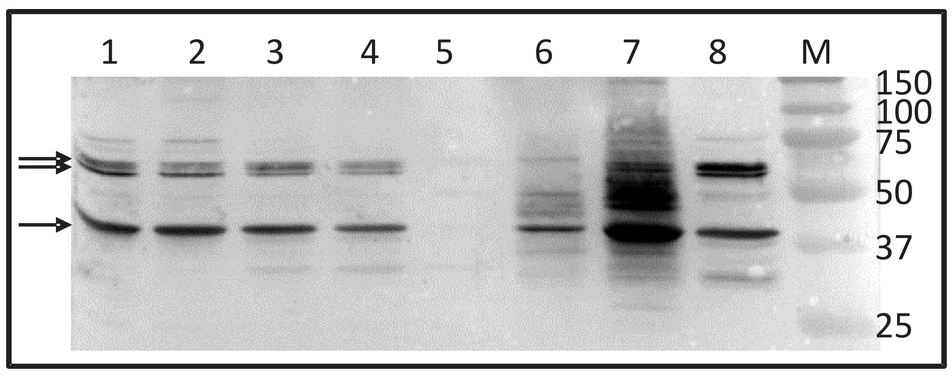
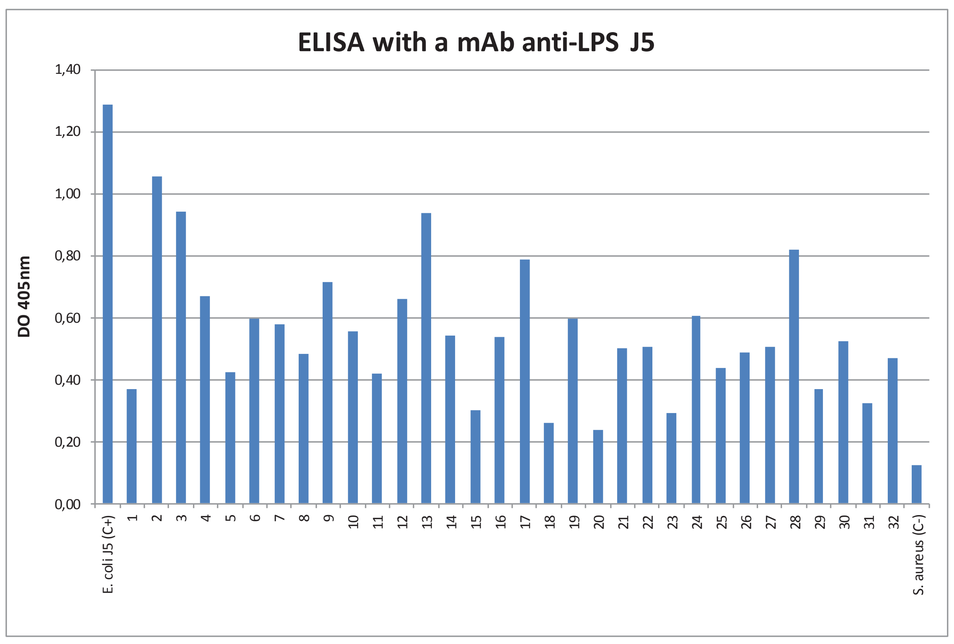


Figure 1. Western blot using serum from a cow vaccinated with STARTVAC®. *S. aureus* cell wall extracts were run in lanes 1-4 (Japanese isolates) and 5-8 (European isolates). M is the molecular weight marker. Numbers on the right indicate the molecular weight of the M (kDa). Arrows on the left indicate the immunodominant proteins in the Japanese extracts that are shared with the European extracts.



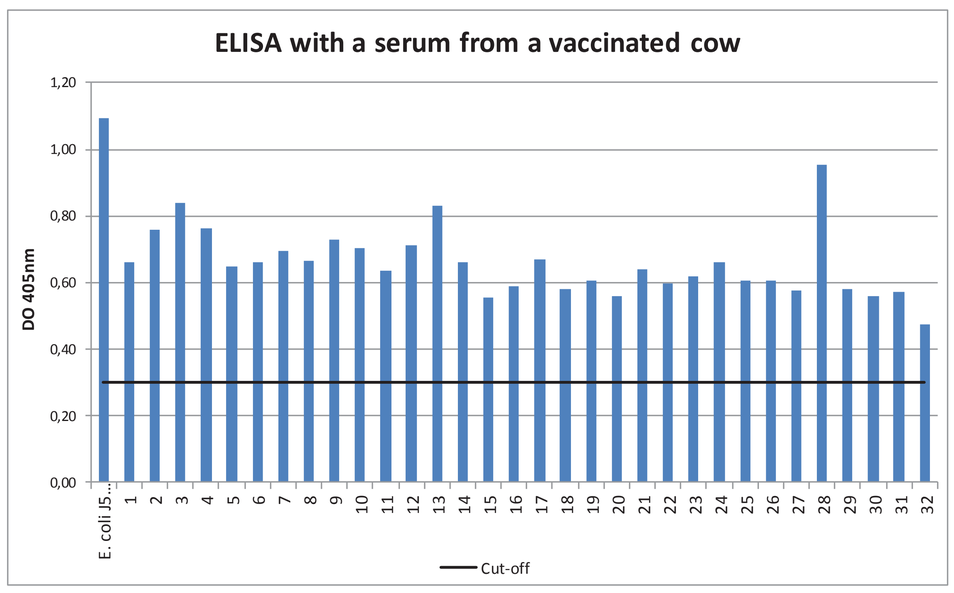


Figure 2. Mean OD in the ELISA with plates coated with European (1-7 columns) and Japanese (8-32 columns) *E. coli* isolates, using a monoclonal anti-LPS J5 antibody or a serum from a cow vaccinated with STARTVAC®.

CONCLUSIONS

This study indicates that *S. aureus* and *E. coli* bovine mastitis isolates from Japan and Europe, present common surface antigens recognized by antibodies induced with STARTVAC® vaccination.

REFERENCES

[1] Prenafeta A, March R, Foix A, Casals I, Costa LL. Vet Immunol Immunopathol 134:208-17. [2] Cole JN, Djordjevic SP, Walker M.J. Methods Mol Biol 2008;425:295-311.

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