

ANALYSIS OF THE ANTIBODIES INDUCED BY UBAC® VACCINATION

Rosa Collado^{1*}, Cristina Solà¹, Ricard Martín¹, Mercè Roca¹ and Antoni Prenafeta¹

¹ R&D Department, HIPRA, Amer (Girona), Spain

*Corresponding author (rosa.collado@hipra.com)



OBJECTIVES

The aim of the present study was to evaluate the antibody response against UBAC® vaccine antigen, the Biofilm Adhesion Component (BAC), with different adjuvants. Moreover, the ability of the generated antibodies to inhibit biofilm formation and to recognize different *Streptococcus uberis* strains was evaluated *in vitro*.

MATERIALS AND METHODS

Four groups of 7 calves were vaccinated with a fixed concentration of BAC antigen and different adjuvants. Each group was vaccinated with 2 mL by intramuscular route at day 0 and 21. Another group of 5 calves was vaccinated with placebo as a control group. Serum samples were obtained at day 0, 28 and 46 in order to study the antibody levels against BAC antigen by ELISA. The ability to inhibit biofilm formation of the antibodies generated through vaccination with the adjuvant Hipramune® U (UBAC® vaccine) was tested by an *in vitro* microplate assay (Sun et al. 2005) with some modifications. The ability of the same antibodies to recognize different *S. uberis* strains was evaluated by ELISA. Data was compared using the Student t-test (SPSSv.22). Significance was declared at $P \leq 0.05$.

RESULTS

Results showed that vaccines formulated with an oil-based adjuvant and Hipramune® U (UBAC® vaccine) exhibited significantly higher serological response ($P < 0.05$) than the aluminium hydroxide-based vaccines and the control group at the end of the study. Moreover, the group vaccinated with Hipramune® U (UBAC® vaccine) showed higher antibody response than the group containing the oil-based adjuvant (Figure 1).

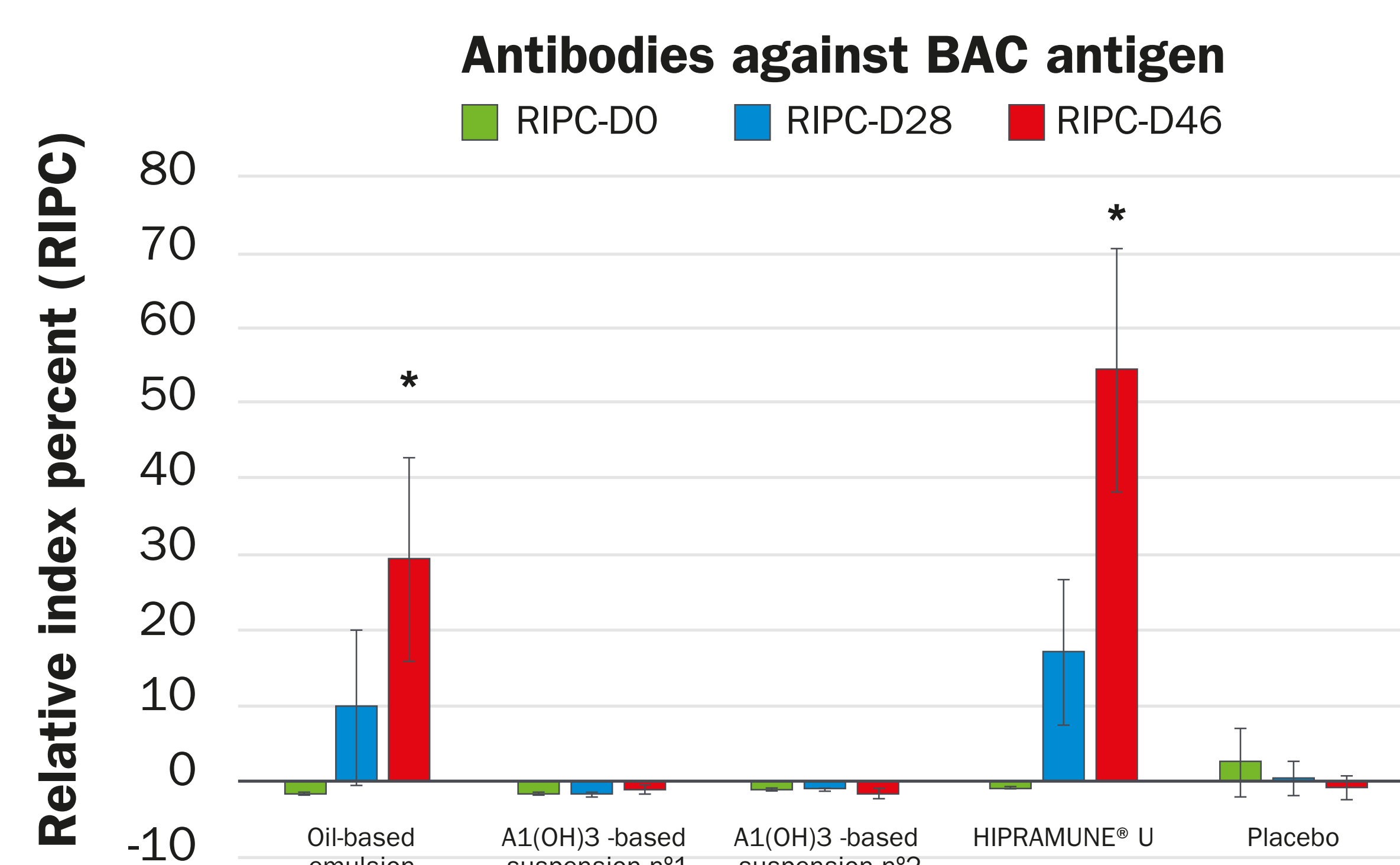


Figure 1. Serological response against BAC antigen combined with different adjuvants.

In the *in vitro* biofilm formation assay, the addition of a 1:2000 dilution of a serum from a vaccinated animal with Hipramune® U or unvaccinated animal to *S. uberis* microplate cultures, did not show any significant difference in their growths (OD_{550nm}) compared to a *S. uberis* culture without serum (Figure 2A).

Nevertheless, when planktonic cells were removed and adhered cells were stained, results showed that wells incubated with serum from a vaccinated animal exhibited lower biofilm formation (OD_{595nm}) than the wells incubated with a serum from an unvaccinated animal or without serum ($P < 0.05$).

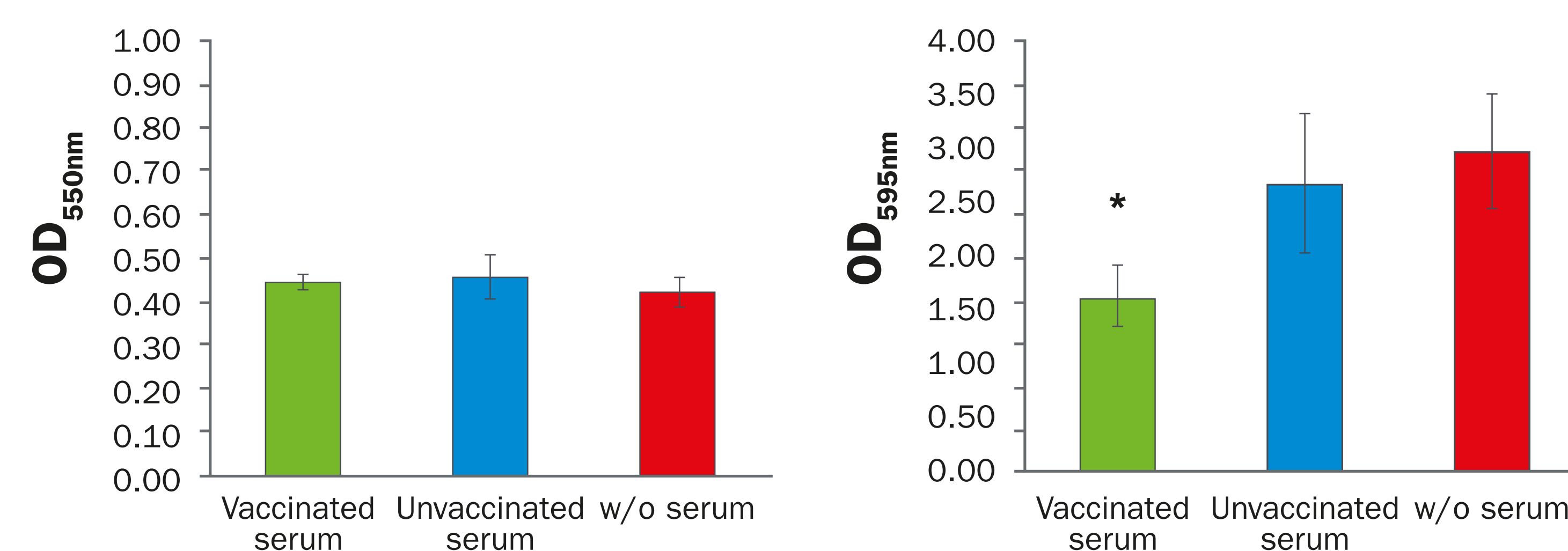


Figure 2. A) Growth of *Streptococcus uberis* SU1H in TSB+YE in presence of a 1:2000 dilution of a vaccinated or an unvaccinated serum and w/o serum. B) Biofilm formation of *S. uberis* SU1H cultures in presence of vaccinated, unvaccinated and w/o serum.

Finally the ability of this serum to recognize BAC antigen from different *S. uberis* strains was tested by ELISA. Results showed that strains from different geographical origin or different biofilm formation ability were all recognized through the vaccinated serum, while a serum from an unvaccinated calf did not. (Figure 3).

Recognition of BAC antigens with a vaccinated and unvaccinated serum

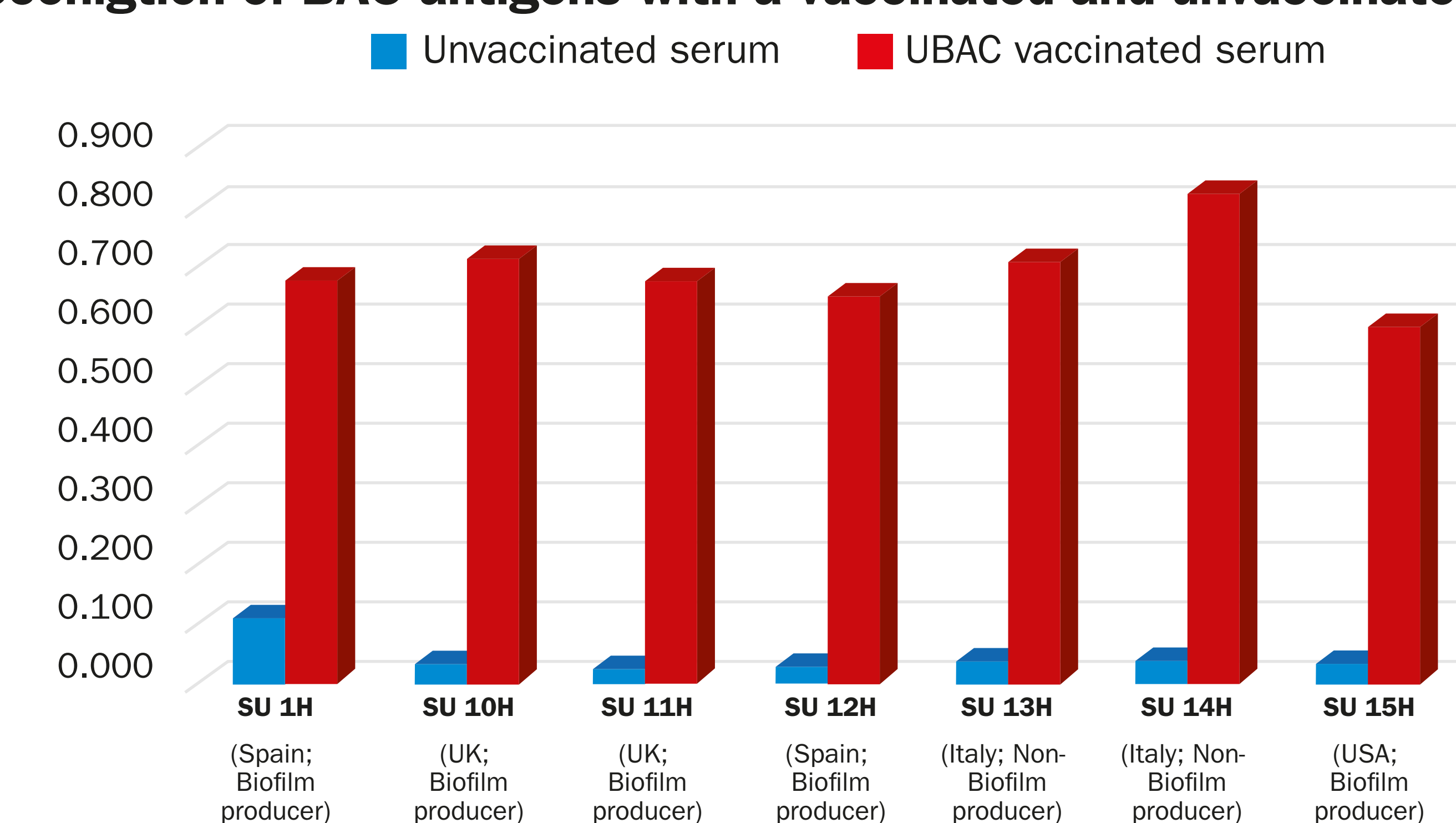


Figure 3. *In vitro* recognition of BAC antigens with a vaccinated and an unvaccinated serum.

CONCLUSIONS

The adjuvant of the UBAC® vaccine (Hipramune® U) induced the greatest antibody levels against BAC antigen compared with other tested adjuvants. Moreover, these antibodies had the ability to reduce the *S. uberis* biofilm formation *in vitro* and to recognize heterologous strains of *S. uberis*, in terms of geographical origin or *in vitro* biofilm formation ability.

REFERENCES

Sun D, Accavitti MA, Bryers JD. Inhibition of biofilm formation by monoclonal antibodies against *Staphylococcus epidermidis* RP62A accumulation-associated protein. Clin Diagn Lab Immunol. 2005 Jan;12(1):93-100.