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Report on multifocal assays for the development of the STARTVAC® vaccine according to a new protocol

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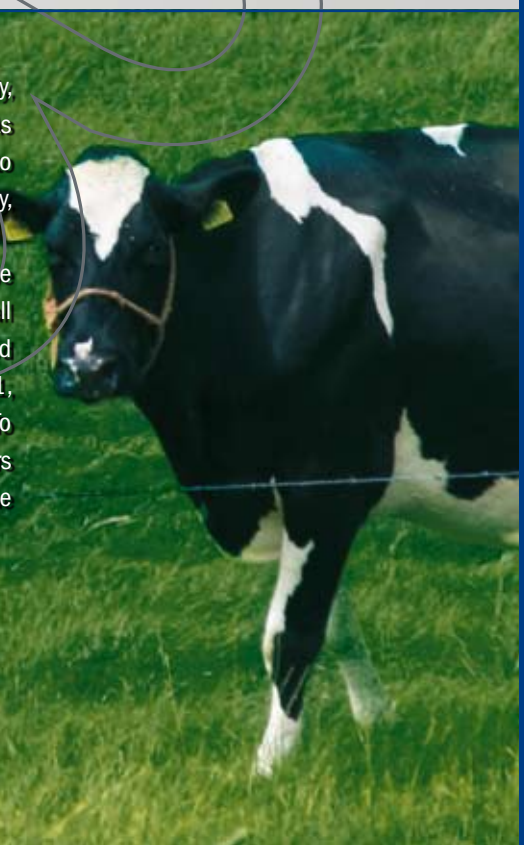
1. Introduction

Vaccination with the STARTVAC® vaccine aims to improve the quality of milk from dairy cow farms. This vaccination induces the production of antibodies against certain biofilm components of *Staphylococcus aureus* and *Staphylococcus spp.* and against LPS, an endotoxin of the cell wall of *Escherichia coli*. This dual effect not only reduces the prevalence of mammary infections caused by *Staphylococcus spp.* but also the severity of the clinical signs which accompany infections caused by *E. coli*.

The MA dossier puts forward a vaccination protocol with three injections given around the dry period of cows (RCP STARTVAC®). The disadvantage of this protocol is that the animals must be vaccinated individually, based on the estimated date of calving. As such, it would be much more practical to vaccinate the entire herd on the same day, with boosters given at regular intervals.

For this reason, we tested an alternative vaccination protocol in a field study: all the animals from a herd were vaccinated at the same time on D1, D21 and D111, regardless of their physiological stage. To maintain this level of protection, boosters were given every 3 months following the three initial injections.



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2. Materials and Methods

Dairy farms (n=10, between 40 and 100 dairy cows, and production levels between 8,000 and 14,000 kg, a total of 531 lactating cattle) of a good technical level were included in the study. For these farms the rearing conditions, such as milking facilities and animal housing, are shown in Figure 1.

Dairy cows and heifers were vaccinated for the first time in May-June 2010, with boosters on Day 21 and Day 111. For 3 out of 10 farms it was not possible to vaccinate the future first-calvers at the same time as the cows. Their vaccination protocol began upon their return to the cowshed to prepare for calving.
At the same time as the first vaccination, the milk quality inspection was conducted in order to identify risk factors in each farm before the start of the study. (The results of these inspections are shown in Figure 2).

During the study, new mammary infections were monitored by conducting bacteriological tests on all cows with cell counts greater than 200,000 cells/ml (included in May 2010 and January and May 2011) and of the milk from cows with clinical mastitis. The cell counts of the milk tank were also monitored throughout the study (Figure 3).

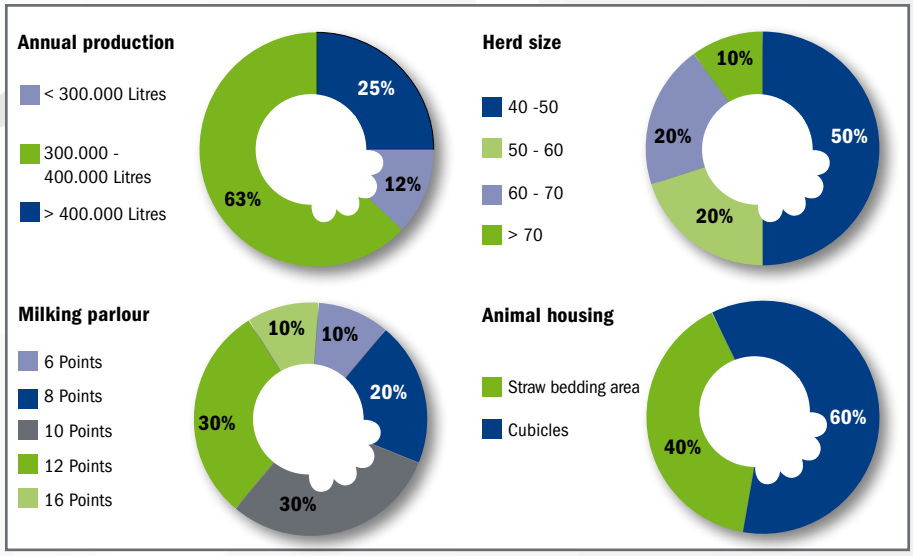

Figure 1. Production, herd size, milking facilities and animal housing of the farms included in the study.

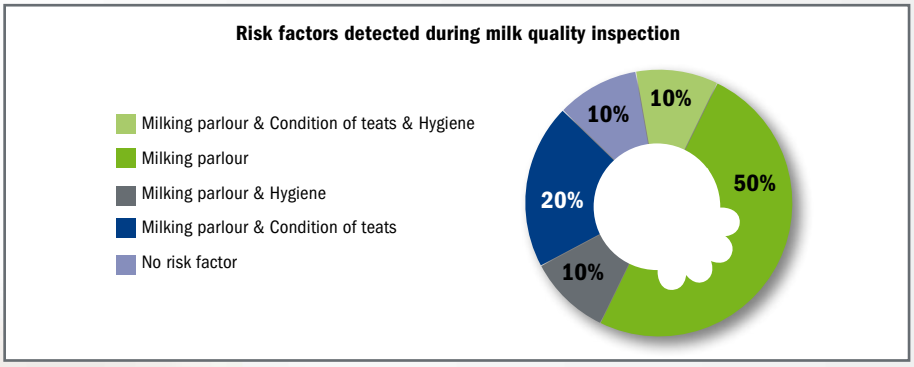

Figure 2. Results of the milk quality inspections made during vaccination.

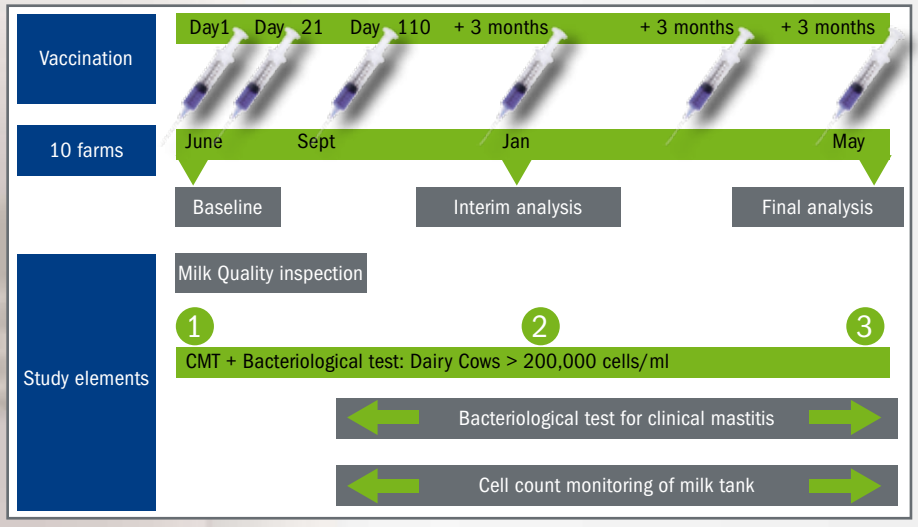

Figure 3 : Study protocol

3. Results and Discussion

In implementing the vaccination protocol, only 2 in 531 cattle showed an increase in temperature (up to 39.6 °C) the day following the injection. The vaccination caused neither pain nor injury at the injection site, nor did it detract from the animals' level of production.

Overall, the percentage of clinical mastitis, as well as the cell counts of the milk tank, decreased from 136% to 55% and from 308,000 to 227,000 cells/ml, respectively, between May 2010 and May 2011 (Figure 4).

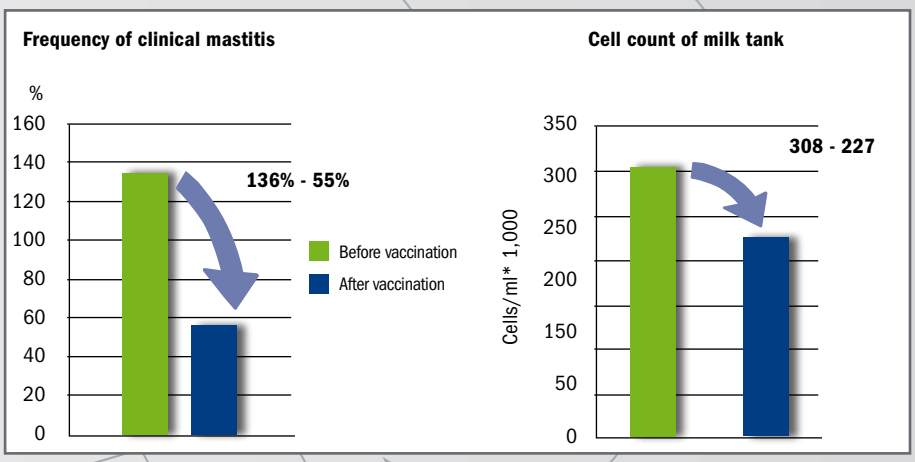

Figure 4. Frequency of mastitis and cell counts of milk tanks on the farms before and after vaccination.

Mastitis rates decreased in 9 out of 10 farms: the remaining cases of mastitis were primarily caused by *Streptococcus uberis* and *Coagulase-negative staphylococci* (CNS) (Figure 5).

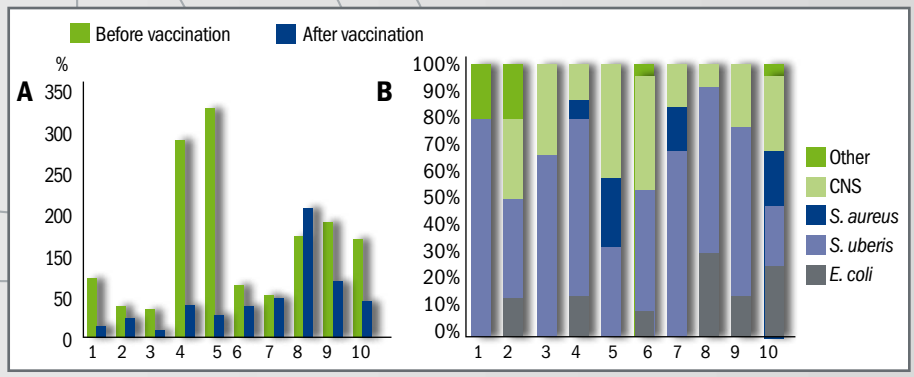

Figure 5. A. Frequency of clinical mastitis before and after vaccination (percentage relative to the number of dairy cows present). **B**. Aetiology of clinical mastitis observed after vaccination with STARTVAC®.

After vaccination, the average cell counts of the milk tank dipped below 250,000 cells/ml in 7 out of 10 farms, compared with only 4 farms before vaccination. Before vaccination, Staphylococcus aureus was found in samples from cows with sub-clinical mastitis in 9 out of 10 farms. After vaccination, Staphylococcus aureus was found in only 3 out of 10 farms (Figure 6).

Sub-clinical infections after vaccination were primarily caused by Coagulase-negative staphylococcal infections. (Figure 6) It is interesting that the decrease in cell counts in the milk tank is not accompanied by a decrease in the number of infected cows, but by a lower number of infected quarters per cow (Figure 7).

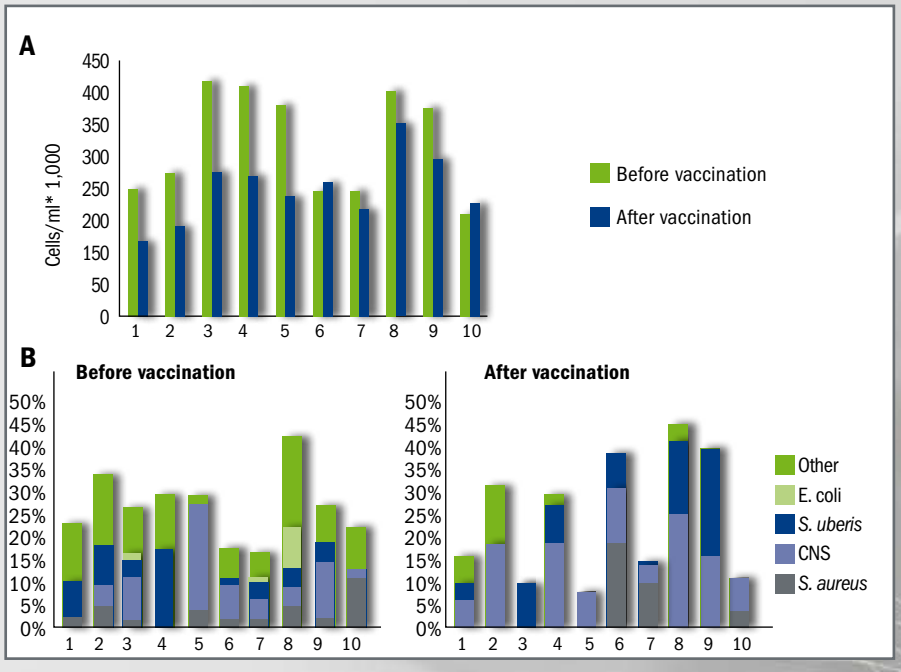

Figure 6. A. Average annual cell counts in the milk tanks of the 10 farms before and after vaccination. **B**. Aetiology of sub-clinical infections before and after vaccination.

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4. Conclusion

Although vaccination with STARTVAC® does not eliminate the presence of E. coli and S. aureus in livestock, the rate of clinical mastitis and cell counts of the milk tank decreased in the majority of the farms. After vaccination, clinical mastitis was primarily caused by *Streptococcus uberis* and *Coagulase-negative staphylococci*, while sub-clinical infections were caused by *Coagulase-negative staphylococci*.

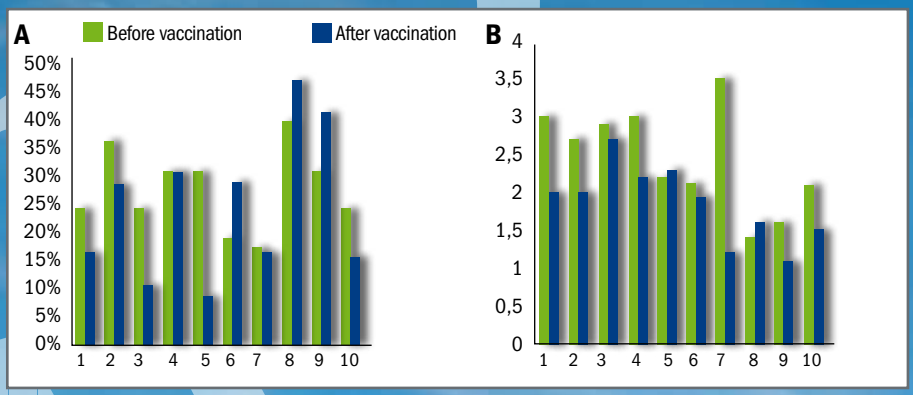

Figure 7. A. Percentage of cows with sub-clinical infections (> 200,000 cells/ml) at baseline and in January 2011. **B**. Number of infected quarters per cow (positive California Mastitis Test) at baseline and in January 2011.