



Monitoring milk for recombinant bovine somatotropin (ab)use

Nathalie G.E. Smits, Susann K.J. Ludwig, Michel W.F. Nielen & Leendert A. van Ginkel

Background

Recombinant bovine somatotropin (rbST) is licensed for enhancing milk production in dairy cows in some countries, for instance the United States, but banned in Europe. RbST use induces antibody formation which, via the bloodstream, are excreted into the milk. Detection of these antibodies can be an adequate approach to discriminate between rbST treated and untreated dairy cows.

Objective

- The development of reliable screening assays for rbST induced antibodies in milk.
- Comparisons of results obtained for both serum and milk samples from a controlled rbST treatment animal trial.
- Comparison of luminex and dipstick format.

Methods

Luminex

- Serum samples: 80x diluted.
- Milk samples: 18x diluted.
- Read out 2 hours after adding the colour-encoded microspheres to the sample.

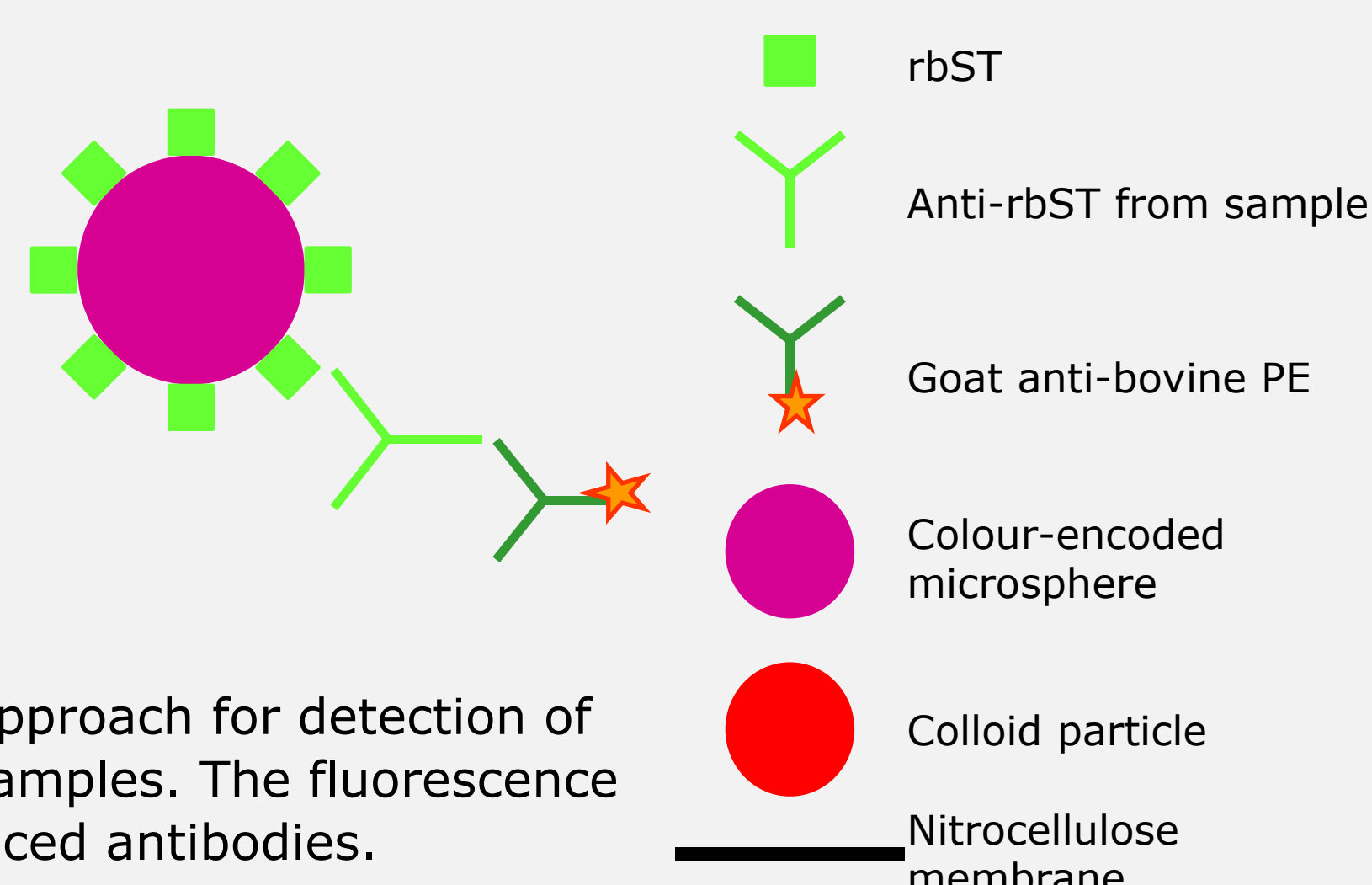


Figure 1. Luminex format using an indirect approach for detection of antibodies against rbST in serum and milk samples. The fluorescence intensity indicates the presence of rbST induced antibodies.

Conclusions

- Responses of rbST induced antibodies in milk correlate to the responses in serum.
- For Luminex, rbST treatment can best be detected in serum.
- For the dipstick further developments are required to obtain an increased sensitivity similar to Luminex.

Literature

Ludwig S.K.J., Smits N.G.E., Bremer M.G.E.G., Nielen M.W.F. (2012). Monitoring milk for antibodies against recombinant bovine somatotropin using a microsphere immunoassay-based biomarker approach. *Food Control* 26:68-72.

Dipstick

- Serum samples: 10x diluted.
- Milk samples: 2x diluted.
- Read out 10 minutes after application on dipstick.
- Prototype dipsticks were kindly provided by Operon.

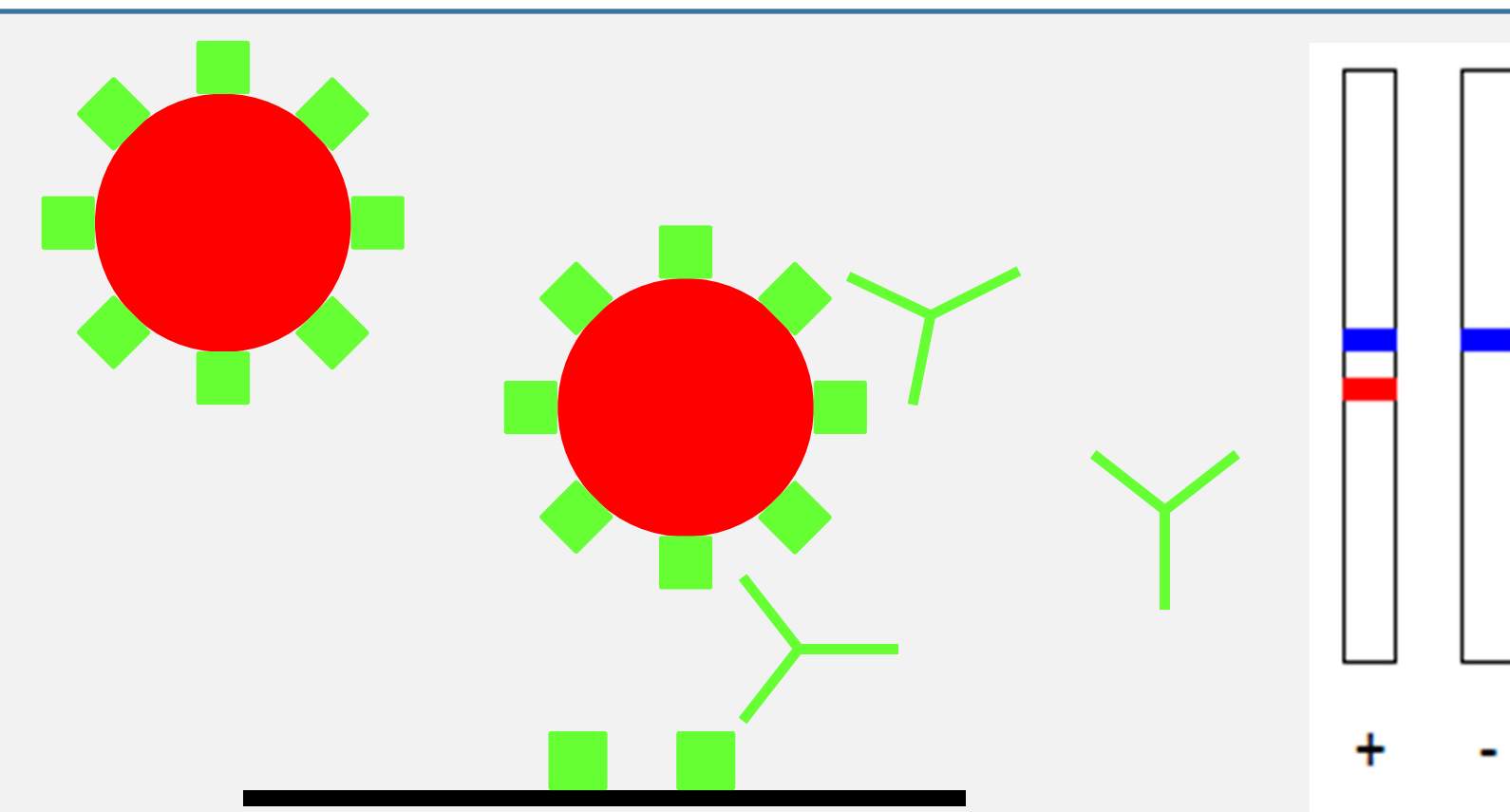


Figure 2. Dipstick format using a sandwich approach for detection of antibodies against rbST in serum and milk samples. A red test line in combination with a blue control line indicates a positive result, whereas only a blue control line indicates a negative result.

Results

Milk and serum samples from different time points of rbST treated and untreated cows were tested. In milk, an antibody response was seen in 67% of the rbST treated cows (Fig. 3A) whereas 94% of the untreated animals did not show a response (Fig. 3B). Moreover, antibody presence in milk correlates with serum antibody response as shown in Fig. 4. But, this figure also shows that presence of rbST induced antibodies in treated animals can be best detected in serum over milk with this method.

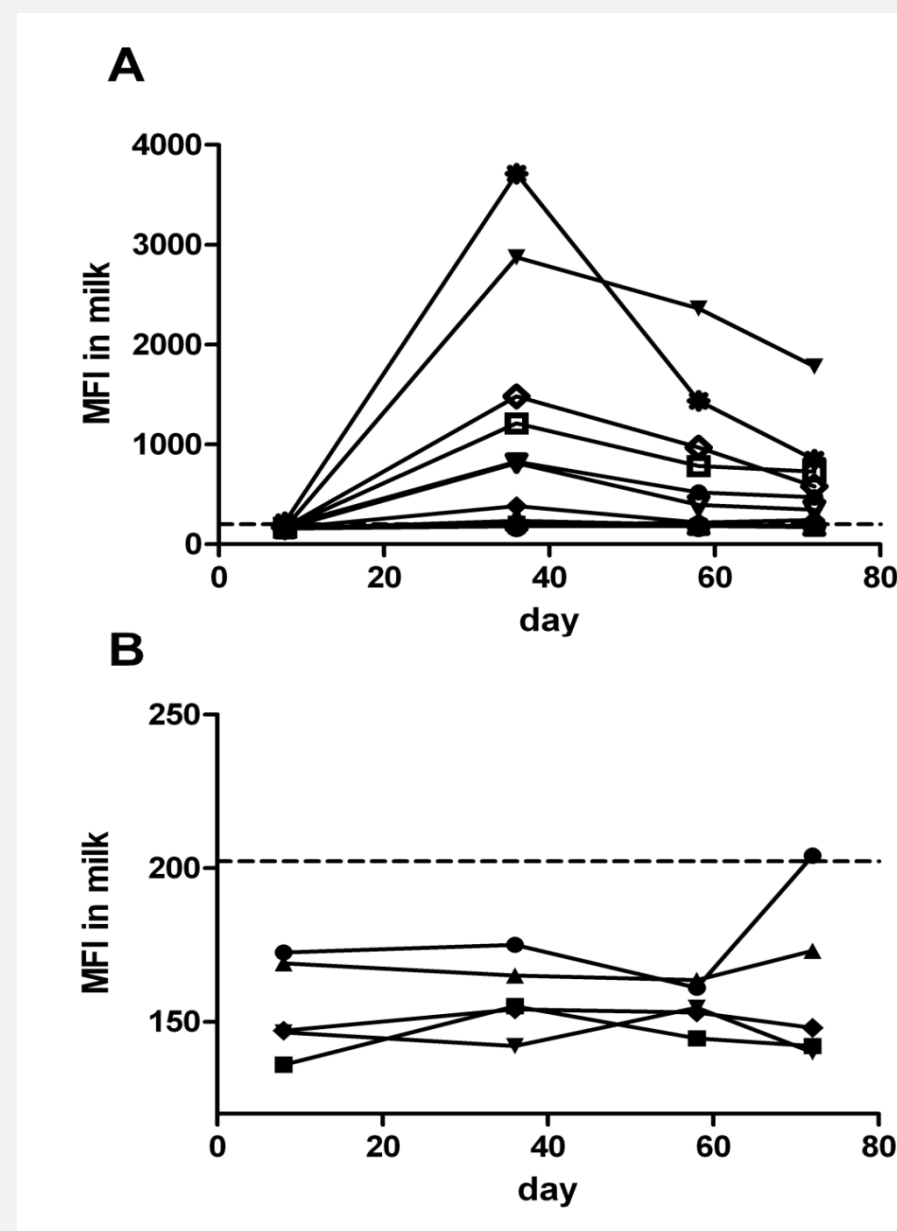


Figure 3. MFI responses at different time points in milk of A) rbST treated animals and B) untreated animals. Dashed line indicates the absolute decision limit.

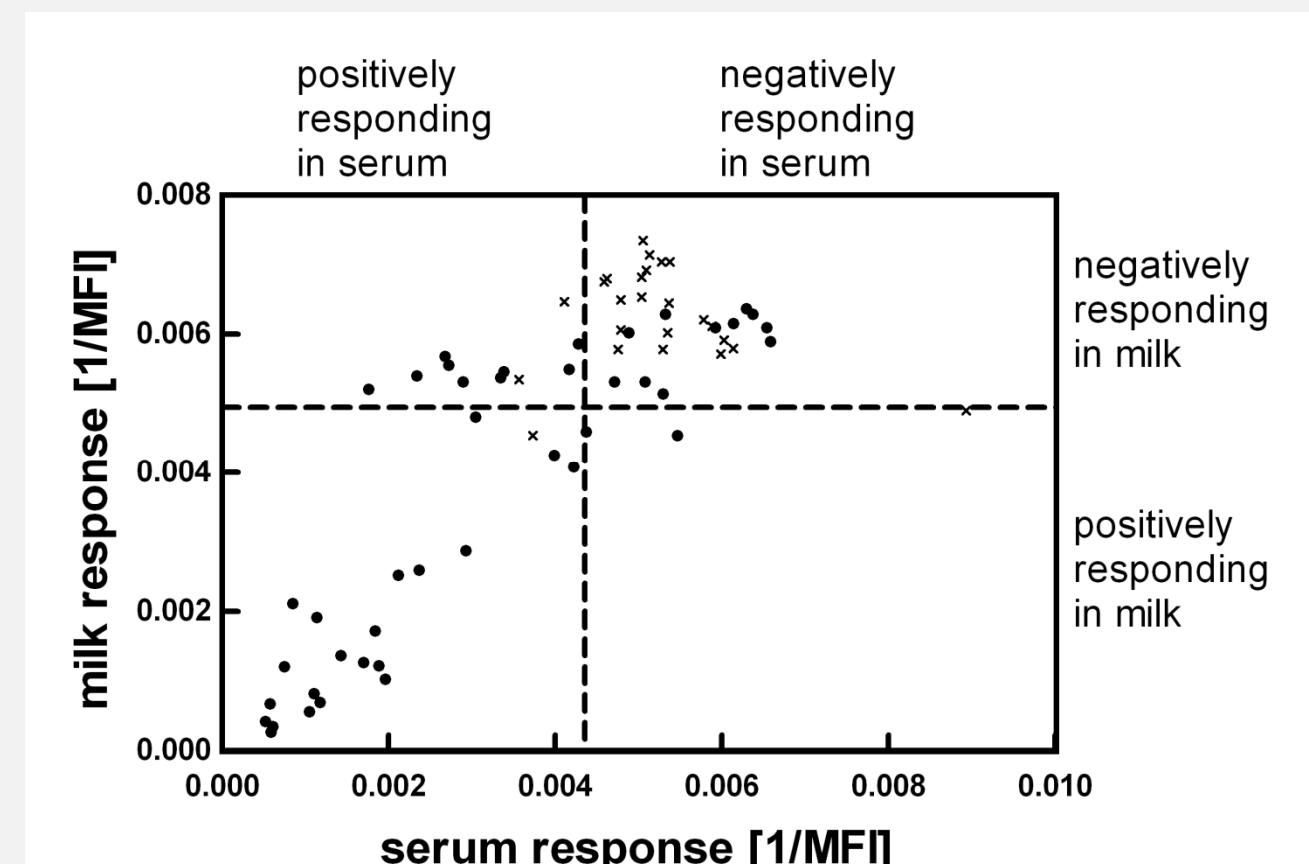


Figure 4. Graphical presentation showing the correlation between anti-rbST responses in milk and serum of rbST treated (●) and untreated (x) animals. Dashed lines indicate the absolute decision limits in milk and serum.

Examples of dipstick results of milk and serum samples from rbST treated and untreated animals are shown in Fig. 5. In milk, an antibody response was seen in 50% of the rbST treated animals where in 95% of the untreated animals no response was detected. For serum rbST treatment was detected in 35% of the treated animals.

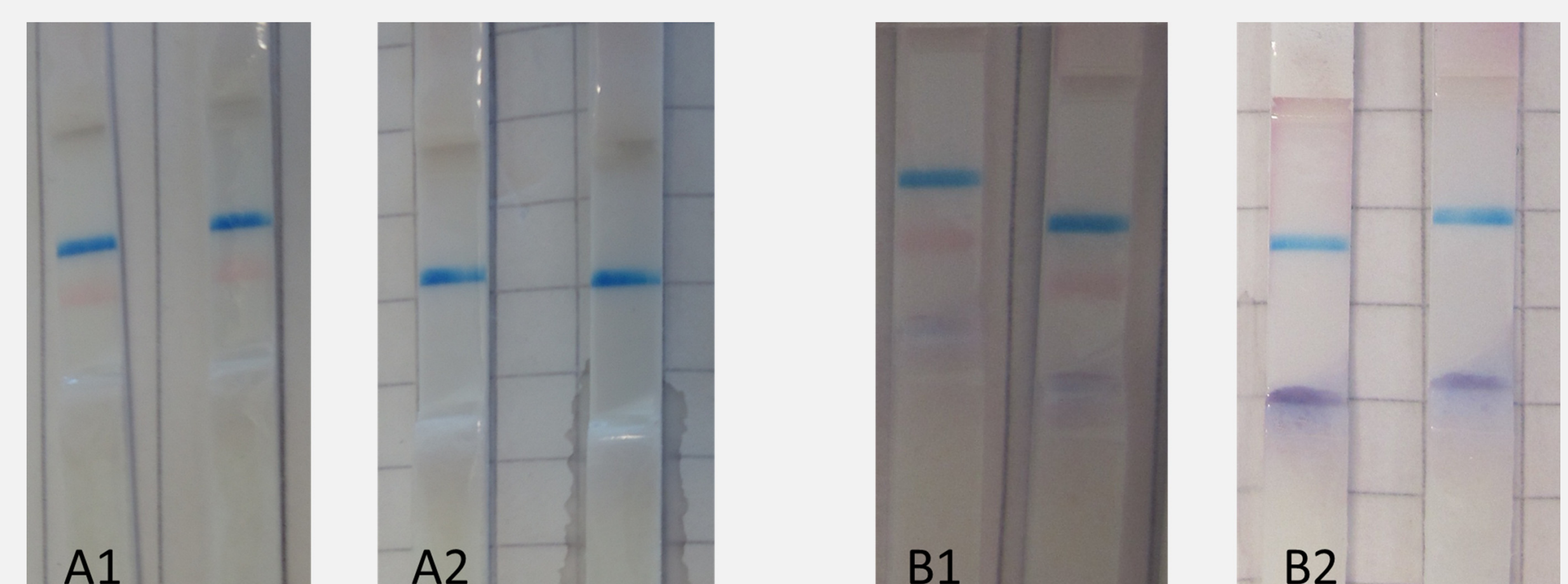


Figure 5. Dipstick results of A) 10 times diluted serum and B) 2-times diluted milk samples from rbST treated (1) and untreated (2) animals.

Acknowledgements

This project was financially supported by the Dutch Ministry of Economic Affairs (EL&I, project 7202901).