Development of an enzyme-linked immunosorbent assay for the serodiagnosis of ringworm infections in cattle

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BACKGROUND

The zoophilic dermatophyte *Trichophyton verrucosum* is the most common agent of dermatophytosis (commonly known as ringworm) in cattle. Several enzyme-linked immunosorbent assays (ELISAs) have been developed for the evaluation of antibody response in animal dermatophytosis, but only few focused on detecting specific antibodies in cattle ringworm.

OBJECTIVES

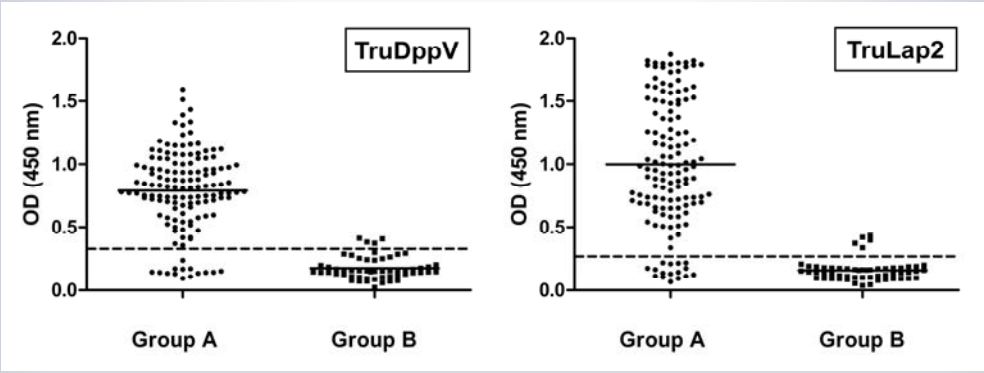
The goal of this study was to develop an in-house ELISA based on recombinant antigens for the serological diagnosis of cattle dermatophytosis.

MATERIALS AND METHODS

Antigens consisted of available recombinant forms of either *Trichophyton rubrum* dipeptidyl peptidase V (TruDppV) or leucin aminopeptidase 2 (TruLap2) which are 98% identical to *Trichophyton verrucosum* orthologues. Sensitivity (Se), specificity (Sp), positive (PPV) and negative (NPV) predictive values of both ELISAs were determined using field serum samples from 135 cattle with ringworm infection confirmed by microscopy and PCR analyses, and from 55 healthy cattle without history of dermatophytosis (negative controls).

RESULTS

Differences between optical density (OD) mean values obtained in both animal groups were highly significant, showing that our ELISAs can discriminate between infected and healthy animals ($P < 0.0001$, Mann-Whitney U test). Using a cut-off point equal to the mean OD + 2 SD of sera from control group, the ELISA detecting specific antibodies against DppV gave 89.6% Se, 92.7% Sp, 96.8% PPV and 78.4% NPV. The recombinant TruLap2-based ELISA displayed 88.1% Se, 90.9% Sp, 95.9% PPV and 75.7% NPV.



Distribution of optical density (OD) values obtained from the sera of cattle with confirmed ringworm infection (group A) and healthy control cattle (group B) using the recombinant proteases *Trichophyton rubrum* DppV (TruDppV) and Lap2 (TruLap2) as coating antigens.
For the TruDppV-based ELISA, the mean ODs of sera from groups A and B were 0.791 0.32 and 0.173 0.089, respectively, and the cutoff value obtained as the mean OD + 2 SD of sera from group B was 0.353. For the TruLap2-based ELISA, the mean ODs of sera from groups A and B were 0.994 0.511 and 0.157 0.085, respectively, while the cutoff point was fixed at 0.327 as described above. With both antigens, a highly significant difference ($P < 0.0001$, Mann-Whitney U test) was noted between OD values obtained when sera from group A versus those from group B were tested. The horizontal bars represent the mean ODs, and the dashed bars represent the cutoff.

CONCLUSIONS

This are to the best of our knowledge the first ELISAs based on recombinant antigens assessing the immune response in ringworm of cattle, being particularly suitable for epidemiological studies and also for the evaluation of vaccines and/or vaccination procedures.

SOURCE OF FOUNDING

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