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Use of a simplified method to determine the avidity of an IgG antibody response against the p38 antigen of *Neospora caninum* in cattle

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Abstract

The detection of specific antibodies in serum from an animal is indicative for an infection with *N. caninum*. However, as e.g. the concentration of specific antibodies fluctuate during pregnancy in persistently infected animals, antibody levels or titres can not be used to discriminate between acute and chronically infected animals. For this purpose, avidity ELISA tests can be used, based on the fact that the first antibodies synthesized after an antigenic challenge have lower affinity for the antigen than those produced later on. Avidity assays available to examine *N. caninum* infections are currently based on end-point titration of specific IgG. In the p38-avidity-ELISA each individual serum is diluted two-fold and at least 8 of these dilutions are tested in both, antigen-coated test and control wells. After the serum incubation, low avidity antibodies are eluted from the test wells by using 8 M urea. This approach guarantees an excellent sensitivity and specificity but it is relatively laborious and reagent consuming.

Using a panel of positive sera from a herd with a recent *N. caninum*-associated abortion outbreak we tried to develop a regression model to estimate the avidity indices for each serum by using only a single dilution. It became obvious that this procedure is affected by the concentration of the specific IgG in each serum. Therefore, a further model was developed to determine from the ELISA indices obtained during the initial screening for positive sera the appropriate serum dilution for the analysis using the single-dilution-p38-avidity-ELISA.