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**Production and Evaluation of Anti-BP26 Monoclonal Antibodies in Serological Detection of Animal Brucellosis**

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**Abstract:**

**Background/Objective:** Brucella BP26 is a good immunogenic antigen and showed high specificity in detecting brucellosis. In China, an Bp26 deleted vaccine M5ΔBP26 has been authorized for use in preventing small ruminant brucellosis, so developing effective detection methods targeting BP26 will be very useful for DIVA.

**Methods:** Using traditional mouse hybridoma technique, we obtained twelve monoclonal antibodies (mAbs) targeting BP26. With a collection of brucellosis positive sera, we evaluated these mAbs’ capacity in detecting different animal brucellosis based on competitive ELISA method.

**Results:** Of the twelve mAbs, only E10 mAb demonstrated sufficient efficiency in detecting brucellosis, as it can be inhibited by 100%, 97.62% and 100% brucellosis positive sera collected from cattle, small ruminants and canine respectively. The E10-based cELISA showed higher accuracy than BP26-based iELISA for cattle and small ruminant brucellosis, especially for small ruminant the sensitivity of cELISA was 97.62% while for iELISA it was only 64.29%. High accuracy was also observed for canine brucellosis, except that the specificity of cELISA was a bit lower than iELISA. The epitope of mAb E10 lies in the amino acid sequence of QPIYVYPDDKNNLKEPTITGY, implying that this sequence can be used as diagnostic antigen for Brucellosis.

**Conclusion:** In conclusion, the E10-based cELISA offers an effective method for detecting the animal brucellosis, which is also crucial for DIVA diagnosis in China where an BP26 mutant vaccine has been massively used.

**Keywords:** brucellosis; competitive ELISA; BP26; diagnosis; monoclonal antibodies