

Rapid detection of antibodies against bovine leukemia virus by bacterial surface complementation assay

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Enzootic Bovine Leukosis (EBL) is a cancerous disease caused by Bovine leukemia virus (BLV). It accounts for considerable economic losses to the dairy industry. The most common and reliable method to diagnose BLV infection is ELISA. However, a major limitation of ELISA is the requirement of manual handling of samples and the long processing time which makes it unsuitable as an on-farm diagnostic test. Hence, there is a need for an alternative test with rapid detection and reduced manual labour. In this regard, antigen-specific antibodies can be detected with a novel biosensor, based on a novel split trehalase (TreA) enzyme, developed in Dr. De Buck's lab. In the current study, the existing biosensor platform was modified to detect BLV antibodies in clinical serum by using bacterial surface complementation of fusion proteins expressed on the outer membrane of *E.coli*. The integration of bacterial surface display system in biosensor format is cost-effective and enhances the speed and sensitivity of the assay. The cells expressing these fusion proteins when incubated along with diluted bovine clinical serum and trehalose (acting as enzymatic substrate) results in production of glucose which can be measured by a commercial glucometer or a visual assay. To validate the assay, a panel of 17 characterized serum samples obtained from BLV positive and negative cattle were tested in comparison with ELISA results. Evaluation of this panel resulted in positive detection of all BLV positive serum. However, 6 false positives were observed among the healthy samples due to the presence of anti-*E. coli* antibodies in the serum samples. Our results indicate that, using this assay, it is possible to detect anti-BLV antibodies in clinical serum and distinguish between healthy and BLV positive samples. In conclusion, the BLV assay is a potential tool for simple and rapid diagnosis of BLV infection which is compatible with both lab-based and a more user-friendly on-farm format.