EVALUATION OF BLOOD SAMPLES COLLECTED ON FLINDERS TECHNOLOGY ASSOCIATES (FTA) CARDS FOR DETECTION OF ANTI-AFRICAN HORSE SICKNESS VIRUS (AHSV) ANTIBODIES USING I-ELISA

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Introduction

African Horse Sickness (AHS) is a highly infectious, non-contagious disease affecting all species of equidae, characterised by high morbidity, mortality and economic losses. The causal agent, AHS virus (AHSV), is endemic in sub-Saharan Africa and 9 distinct serotypes are recognised (Fig 1). Laboratory confirmation of disease is imperative for implementation of control measures. Integral to laboratory diagnosis is choosing appropriate specimens, correct sampling and safe transportation to the laboratory to maintain biosecurity and sample integrity. FTA cards have been used for the safe collection of blood for detection of nucleic acids and antibodies against several viruses, but there is a lack of information about their use in AHS diagnosis.

Project objectives

1. Determine safety of transportation of AHSV infected blood on FTA cards and antibody detection in card eluents using I-ELISA
2. Validate the I-ELISA for detection of anti-AHSV antibodies in FTA card extracts using the serum based I-ELISA as the golden standard.
3. Corresponding experimental (n = 10 positive and 10 negative each) and field (n = 30 positive and 97 negative each) blood and sera were used in the exercise.

Milestones/achievements

- Live virus could not be isolated from all FTA card extracts, proving safety of transportation of blood samples on FTA cards
- Viral nucleic acids were detected from extracts of FTA cards blotted with AHSV positive blood, thus enabling molecular downstream processes
- Anti-AHSV antibody positive and negative results were obtained from extracts of positive and negative bloods accordingly. Analytical and diagnostic sensitivities and specificities of 100% were demonstrated (P>0.05) and agreement between results was moderate to excellent (Fig 2)
- FTA card eluent based I-ELISA demonstrated good reproducibility and repeatability (coefficient of variability <20%)

Conclusions

Blotting of blood on FTA cards provides a safer alternative to transportation of the infectious biological fluids in vacutainer tubes which may break and leak. FTA card eluents can be used for the serological diagnosis of AHSV infection using I-ELISA, and the assay's diagnostic specificity can be optimised by storing the cards at –20^{0}C or below.

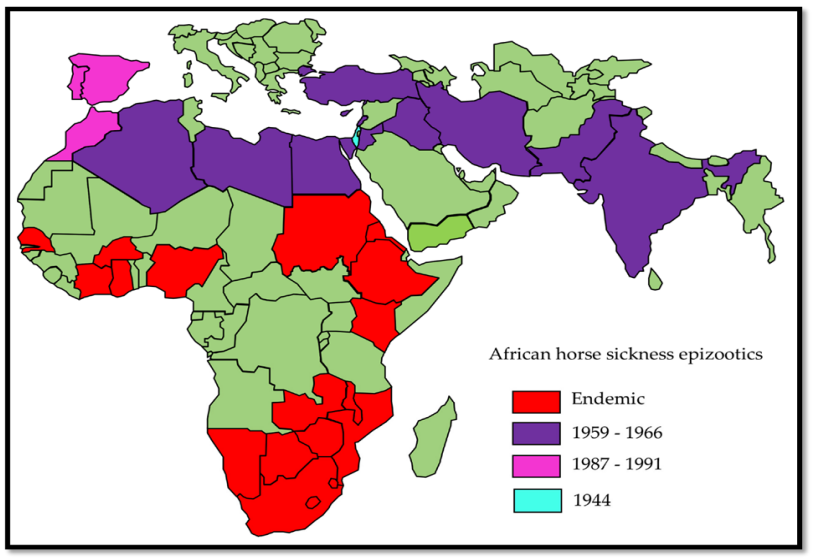
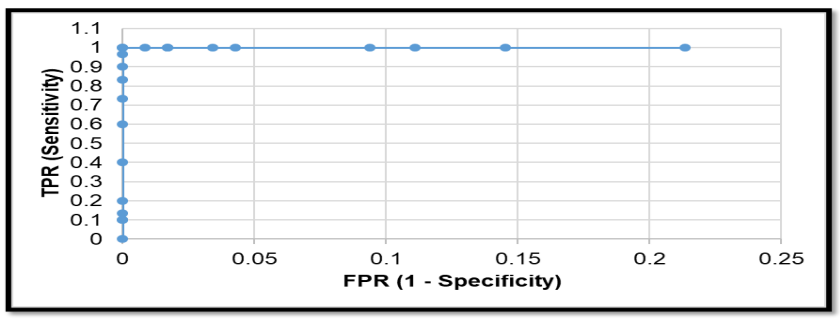


Fig 1 A map of AHS outbreaks that occurred worldwide during the last century (Dennis *et al.,* 2019)



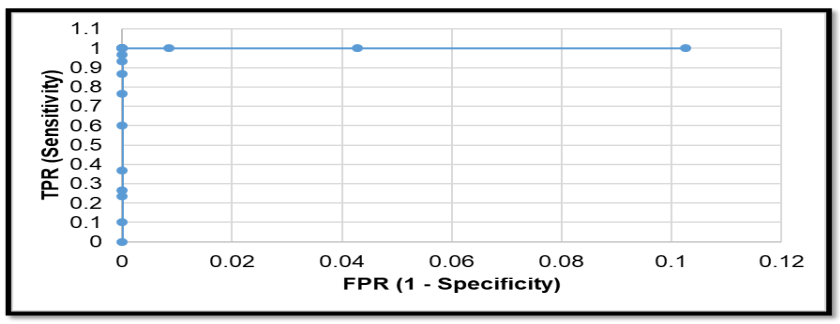


Fig 2 Agreement between the performance of the sera (top) and FTA card extract (bottom) based I-ELISA depicted by respective receiver operating characteristic curve data

Other

The FTA card sampling technique is a feasible alternative to the classical bleeding of horses in vacutainer tubes for sample collection intended for AHSV antibody detection using I-ELISA.

Future Plans

To organise an FTA card sample based AHS serological proficiency test scheme, where the samples would be tested by different analysts (inter-analyst) using different equipment and methods, as part of continued validation for the method.

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