TrichLabCheck – A voluntary trichomonosis inter laboratory comparison project

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TrichLabCheck – A Voluntary Trichomonosis Inter Laboratory Comparison Project In South Africa

Industry Sector: Cattle And Small Stock

Research Focus Area:

- **Animal Health and Welfare**

Research Institute: University Of Pretoria

Researcher: Dietmar Holm

Research Team

Title	Initials	Surname	Highest Qualification	Research Institution
Dr	T	Zangure	BVSc	University of Pretoria

Completion: 2020

Aims Of The Project

- This study aimed to validate the accuracy of voluntarily enrolled private (n = 8) and state-owned (n = 5) laboratories that perform trichomonosis diagnostic tests by estimating the sensitivity (Se) and specificity (Sp) per laboratory. It was hypothesized that diagnostic laboratories in South Africa play an insignificant role in the inaccuracy of the diagnosis of trichomonosis.

Executive Summary

Trichomonosis is currently the most important venereal disease of cattle in South Africa with adverse economic implications to the beef production industry due to cow abortions, infertility and culling of carrier bulls. Once diagnosed in a herd, eradication is difficult due to financial and biological implications. Bulls are asymptomatic carriers and susceptibility increases with age. In infected females, clinical signs include embryonal death, abortion, pyometra, foetal maceration and uterine discharge.

Diagnostic accuracy is one of the major clinical problems preventing easy eradication of trichomonosis from a herd and can be influenced by biological variance in the occurrence of the organism, sampling errors, sample degradation during sample transport and diagnostic laboratory inaccuracies.

Objective Statement

The objective of the project was to determine the role that diagnostic laboratories play in the inaccuracies of trichomonosis diagnosis in South Africa.

Results

Laboratories performed either the culture method (n = 5), polymerase chain reaction (PCR) (n = 6) or a combination of culture and PCR (n= 2). Fresh preputial scrapings from four bulls with known negative status for trichomonosis were pooled in 200ml of phosphate buffered saline (PBS) to form the sample base for 12 subsamples of 13ml each. Duplicate subsamples were then contaminated with 2ml originating from four different laboratory cultures of *Tritrichomonas foetus* or 2ml of culture medium for four negative samples. Aliquots of the subsamples were transferred to an anaerobic transport medium, and the final concentration reached in these samples submitted to the laboratories, were categorised as follows: weak (30 organisms/µl). A total of 312 samples were sent by courier in two separate rounds: eight (4 duplicates) positive and four negative samples per round. Multiple logistic regression was performed on sensitivity, using sampling round, laboratory sector, diagnostic test type and sample concentration as independent variables, and removing variables in a stepwise manner based on the highest P-value.

Two public laboratories only reported on one round of sampling, and one batch of 12 samples was severely delayed in reaching another public laboratory. The sample identifications of a further two batches were not recorded by the respective private laboratories. The results from these 60 unreported samples were not included in the analysis. Laboratories that performed the PCR assay (solely, or in addition to culture) were grouped for data analysis. The overall specificity (Sp) was 100% and the sensitivity was 88.7% (95% CI 83.9% – 93.5%). Laboratories using PCR recorded higher sensitivity than those using the culture method (95.5%; 95% CI 91.0% – 99.9% and 81.3%; 95% CI 72.5% – 90.0% respectively, $P < 0.01$), and private laboratories recorded higher Se than public laboratories (96.4%; 95% CI 92.9% – 99.9% and 73.2%; 95% CI 61.2% – 85.2%, $P < 0.01$). For laboratories using PCR, weak positive samples recorded a lower sensitivity than strong positive samples (86.4%; 95% CI 70.8% – 101.9% and 100%; 95% CI 100% – 100%, respectively, $P < 0.01$). One public and six private laboratories obtained 100% accuracy during the two sampling rounds.

In the logistic regression model, private sector (compared to public), an increasing concentration of organisms in the sample and the second round of sampling (compared to the first round) were independent predictors of laboratory sensitivity for the detection of *Tritrichomonas foetus*.

Conclusion

It is concluded that inaccuracies in the diagnostic laboratory contributes to the deficiencies in diagnostic sensitivity for trichomonosis in South Africa, but does not influence diagnostic specificity. It is further concluded that diagnostic sensitivity was independently influenced by the sector in which the laboratory operates (private vs public) and the concentration of Tritrichomonas foetus organisms in the sample.

POPULAR ARTICLE

Trichomonosis: What Role Does The Laboratory Play In Combating The Disease?

Prof Dietmar Holm

INTRODUCTION

Trichomonosis is a venereal disease of cattle that results in significant losses to the beef industry in particular, due to a severe reduction in the reproductive potential of beef herds. The disease occurs worldwide and is currently widespread in South Africa. In many cases herds are infected without the knowledge of the farmer, because there are often no external signs visible in the cattle. This means that without knowing, farmers loose thousands of rands in potential income due to the loss of unborn calves.

The diagnosis of trichomonosis is done on bulls, and must be performed by a qualified veterinarian only. Incorrect sampling results in incorrect diagnosis, which means that a farmer will remain in the dark about the status of his or her cattle herd. After collecting samples, they are submitted to a laboratory for diagnosis. This diagnosis can be done using different types of tests and must also be done under strict controlled conditions. Several laboratories in South Africa perform this service for veterinarians.

The nature of the disease is such that the diagnostic test is not 100% accurate, even when done by the correct professionals. In a recent study performed by the Faculty of Veterinary Science at the University of Pretoria, the role of diagnostic laboratories in the accuracy of the diagnostic test for trichomonosis was investigated. The TrichLabCheck research team, led by Prof Dietmar Holm, found that indeed in South Africa, amongst the 13 laboratories that voluntarily participated in the research, several false negative

results were reported. There were no false positive results reported in this study to date, which is in line with similar studies done elsewhere in the world. This is important information for veterinarians and farmers in South Africa, who need to consider that in some cases a bull that tested negative may in actual fact be positive and needs to be tested again to confirm his negative status. It was found in this study that the average diagnostic sensitivity of all participating laboratories to detect trichomonosis was 88.7%. This means that potentially for every 10 positive bulls tested in South Africa, at least 1 will provide a false negative test result.

The research emphasises the need to perform repeated samples on individual bulls to confirm their individual negative status, or to test a large number of bulls in a given herd to confirm the negative status of a herd. It also highlights the fact that a negative test result of a single bull in a positive herd must be interpreted with care, because it may just be that the particular bull gave a negative test result when in fact he may be infected with the disease.

“Trichomonosis has been an increasing problem in South African beef cattle over the past decades, and we are hoping that farmers and veterinarians will use this research to be more vigilant in their diagnostic approach towards the disease”, prof Holm stated.

The research further confirmed that the number of Trichomonas organisms in the sample contributes to the accuracy of the test. This emphasises the importance of using only a qualified veterinarian to perform this important task on farms. Dr Tinashe Zangure, the masters student and veterinarian involved in this study confirmed that he gained excellent knowledge not only about the disease, but also about the importance that bull sampling and laboratory techniques play in the effort to combat the disease in cattle herds.

The University of Pretoria, in collaboration with the Ruminant Veterinary Association of South Africa, will soon be publishing a list of laboratories with acceptable levels of accuracy for trichomonosis. The research will be ongoing, and the list will be updated in an effort to ensure that the veterinary industry strive towards diagnostic excellence in South Africa.

Please contact the Primary Researcher on the project if you need a copy of the comprehensive report – dietmar.holm@up.ac.za

Dietmar Holm

Animal Health and Welfare, Cattle and Small Stock

2020, Animal Health & Welfare, CSS, Holm, Online, UP

< STEC from feedlot to abattoir

> Larvicide testing for blackfly control

DEADLINES for RESEARCHERS 2021

Proposals for 2021: TBC
Progress reports: 28 Jan 21
Final reports: 29 Jan 21 Final includes comprehensive report and popular article

COMMITEE MEETINGS for 2021

RMRDSA CSS Planning - TBC
Project Committee - TBC
Pork Planning - TBC

Calendar

< Apr 2021 >

Sun	Mon	Tue	Wed	Tur	Fri	Sat
				1	2	3
4	5	6	7	8	9	10

11	12	13	14	15	16	17
18	19	20	21	22	23	24
25	26	27	28	29	30	

PORK Priority Areas

Cattle & Small Stock Programmes

1 Sustainable natural resource utilisation

2 Improvement of Livestock production and forage

3 Management of agricultural risk to create a resilient Red Meat sector

4 Sustainable health and welfare for the Red Meat sector

5 Enhancement of production and processing of Animal Products

6 Consumer and market development of the Red Meat sector

7 Commercialisation of the emerging sector