### Bovine Trichomoniasis Veterinarian Certification



### Trich—the basics

*Tritrichomonas foetus* is a major cause of fetal wastage wherever natural breeding exists

1932—first reported case of trichomoniasis in U.S. cattle

World-wide prevalence (A few European countries declared Trich free)



## The organism

Protozoa Pyriform shaped Rounded anterior Pointed exterior 3 anterior flagella 1 posterior flagella



Size: 20 X 10 µm (~ size of bovine sperm head)



### Infection

Venereal disease Can ONLY be spread during the breeding act

Carried by male

Positive males, asymptomatic

Negative clinical effects limited to female



## **Clinical signs**



Pyometra: 5% - 15% of the infected females

Abortion: 5-7 months gestation (literature: 30% of cases)

Prolonged calving interval: primary herd sign – peak fetal loss is 50 to 70 days of gestation





Fig. 2. Distribution of the calving percentages by month in *T. foetus* - infected and non-infected herds

The bars represent the means of compiled data corresponding to the calving dates from 419 cows of 32 infected herds and 530 cows in 32 non-infected herds.



## Female immunity

Usually develop short term immunity ~100 days after fetal loss



Immunity lasts 2-6 months Susceptible to re-infection late this breeding season or next

Can become chronic carriers: ~0.3% Carry live calf to term

References listed at the end of the presentation



### Trich vaccination

One currently available TrichGuard™ & TrichGuardV5L™



Mixed efficacy/effectiveness reported in the literature ~66% preg rate vaccinated heifers ~28% preg rate unvaccinated heifers Exposed to

Exposed to experimentally infected bulls

Off-label use in bulls



### Trich vaccination study: 2017

TrichGuard™ Heifers 20 treatment: 20 controls All vaginally inoculated heifers 49 day breeding season Preg rate (25-41 days gestation) vaccinates, 95% controls, 70% Embryonic loss vaccinates, 47% controls, 71% p value: 0.153



### Trich in the male

Male is an <u>asymptomatic</u> carrier



T. foetus is NON-invasive—lives in penile crypts/folds



### Trich treatment

### NONE:

No legal treatment for either males or females

N=1 study Metronidazole successful inhibition of Trich in vitro and in vivo Is illegal to use in food animals in the U.S.



### Risk factors--age

Bulls < 4 years age are less likely to be carriers

Bulls < 4 can become chronic carriers

Bulls  $\geq$  5 years age 2.2X more likely to be positive than bulls 2-5 years of age

Bulls > 3 years age more likely to become infected



### Risk factors--breed, mgmt.

Bos taurus vs. Bos indicus Bos taurus bulls 6X more likely to be infected

Herds with ≥500 cows 13X more likely to be positive

Bull: Cow ratio <1:25 2.2X more likely to be positive



### Risk factors--management

Odds of being a positive herd if neighboring a positive herd was 18.3 (CI: 4.1-81.1) compared to neighboring negative herds

Herds that reported  $\geq$  1 week elapsed time to fix broken fence(s) were 4.3 (CI: 0.9-20.2) time more likely to be positive.



## Risk factors—herd commingling

Odds of being a positive herd if grazing on public land compared to private land with was 2.9 (CI: 0.7-12.1)

Herds commingling with 14 or more herds Risk of infection was 19X higher compared to lesser commingled herds



### Prevalence



Florida: 30.4% operations had at least 1 positive bull 0-27% of the bulls within operations were positive

California: 15.4% operations 4%-38.5% of bulls positive

Oklahoma: 3.0% bulls that were tested Salebarn sampling

Tennessee: <0.01% 1,979 dx test records & 458 slaughter bulls

Australia: no cases since 1988 positive cases found in 2020



### Laboratory Diagnostics



### Trich diagnostic samples

Smegma—best 1.5 to 2.0 ml minimum

Vaginal mucus

Pyometra



## Sampling methods

Gauze sponge used to collect smegma

VS.



Pipette

#### Both methods accepted in Kansas



# Polymerase chain reaction (PCR)

**Identifies DNA** 

Is specific for *T. foetus* 

Enteric non-pathogenic Trich organisms can look like T. foetus on culture



# Polymerase chain reaction (PCR)

Two shipping media options available

1. InPouch™ TF



Media accepted by ALL states that have Trich regulations

Call the state you are shipping bulls to before collection!



# Polymerase chain reaction (PCR)

Two shipping media options available

2. Phosphate buffered saline or 0.9% saline

Media accepted by SOME states: Kansas, Nebraska, South Dakota, Colorado?

Call the state you are shipping bulls to before collection!



### PCR

Samples collected in InPouch<sup>™</sup> TF or InPouch<sup>™</sup> Tube

Samples must be maintained at 59° to 99°F after collection and during transport to KSVDL

Must arrive in the lab within 72 hours of collection





AS



KSVDL research: pouches < 6 months expired are acceptable

If samples submitted in pouches >6 months expired—a disclaimer will be added to the result



### PCR

### Samples collected in PBS/Saline

Samples shipped similar to other diagnostic samples: on ice

No time requirement from collection to arrival to lab No expiration date

KSVDL will ship free tubes for your use (you pay shipping)



### Winter shipments: InPouch™

Unless specify Saturday UPS delivery, do not ship on Friday

Keep samples warm Hand warmers Ice packs heated/microwave

Or incubate and ship on ice (later slide)



## Sample-Pooling



### Sample pooling is available

Each bull's smegma must be put into a separate pouch/tube

The lab will pool the samples

Lab will pool up to 5 bulls in one test sample

Positive pools: each bull in that pool will be individually tested



### Sample-Pooling



#### Not all states accept pooled Trich test results



## Optional Sampling Handling Method—Incubation/freezing

<u>InPouch</u>™

Incubate at clinic at 93-100°F for 43-48 hours

Then freeze

Send to KSVDL on ice



### InPouch<sup>™</sup> TF



Store pouches/tubes horizontally at room temperature away from light

Are biosafety category 2: should be autoclaved or disposed of appropriately

Never store in the refrigerator

Do not use if media is cloudy, dark brown, or dry



### PCR

Analytic Sensitivity: 100 organisms / ml media

#### Diagnostic Sensitivity: 1 test ~ 99.0%

#### Diagnostic Specificity: 1 test ~ 99.9%

References listed at the end of the presentation



### Laboratory variability

Blinded study, inoculated pouches, 20 samples, study repeated twice



Laboratory

### Diagnostic Sensitivity (%)

		Sample		Overall
	Collection	handling	PCR	success
Scenario	success	success	success	(sensitivity)
1	99.9	99.9	99.0	98.9
2	99.9	80.0	99.0	79.1
3	80.0	99.9	99.0	79.1



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