



VetMAX-Gold Trich Detection Kit

The only USDA-licensed, real-time PCR test for the detection of *Trichostrongylus axei* (*T. axei*) in bulls

Features

- First USDA-licensed, *T. axei* real-time PCR detection kit
- Highly sensitive, qualitative real-time PCR assay
- Pool up to 5 samples with 96.16% accuracy
- Proven repeatability and reproducibility

Benefits

Unique test

The Applied Biosystems™ VetMAX™-Gold Trich Detection Kit is a USDA-licensed diagnostic test that has successfully passed the USDA's stringent review process. This process confirms both the effectiveness of our real-time PCR test and the compliance of the production and quality systems in our manufacturing site.

Fast and simple to use

- Simplifies diagnostic lab workflow
- Easily automated for high-throughput processing

Economical

- Detection kit can be used to test pools of up to 5 samples
- Pooling of samples helps reduce cost to producer

Reliable and robust

- Highly sensitive, qualitative real-time PCR assay that has yielded 100% sensitivity in field trials
- Helps improve reproducibility across labs and across states
 - Study performed at three external sites
 - Positive agreement: 96.99%
 - Negative agreement: 98.94%

- Helps improve quality control
 - Applied Biosystems™ *T. axei*-Xeno™ Control DNA Mix
 - Applied Biosystems™ Xeno™ Control DNA
 - Applied Biosystems™ *T. axei* Primer Probes Mix for optimized multiplex PCR amplification of both the Xeno Control DNA and *T. axei* targets

Confidence in results backed by data

A study was conducted of the pooling of cultured samples and comparison of multistate laboratory workflows with the Applied Biosystems™ MagMAX™ sample preparation system and Applied Biosystems™ VetMAX™ qPCR reagents for detection of *Trichostrongylus axei*-colonized bulls.

The objective was to determine the effect of pooling a single positive sample having various C_t ranges with four negative samples (1:5). If a negative effect was seen, a 1:3 pooling study would then be conducted. The goal was to compare different sample preparation systems and various real-time PCR feeder lab workflows with our 5X Applied Biosystems™ MagMAX™ Pathogen RNA/DNA Purification Kit and amplification with VetMAX *T. axei* reagents workflow. The study assessed the specificity of the VetMAX *T. axei* reagents by sequencing all positive samples with C_t values less than 38 and suspect sample C_t values between 38 and less than 40 cycles.

95.6% agreement was reached between the Kansas State Veterinary Diagnostic Laboratory (KSVDL) using MagMAX and VetMAX *T. foetus* reagents and the feeder laboratories. Evaluation noted 1:5 pooling is likely to miss 4% of the positives and 1:3 pooling is likely to miss 3.5% of the positives. 175/176 positive samples were confirmed to be *T. foetus*: one sample could not be sequenced with the primers designed for this study.

Bovine trichomonosis

Bovine trichomonosis is a sexually transmitted infection caused by *T. foetus*, which is a flagellated protozoan parasite. It colonizes in the vaginal, uterine, oviduct and preputial epithelium and results in embryonic death, abortion and infertility in the female. Bulls are the main carriers of *T. foetus*, remaining asymptomatic for their entire lives, and there is no treatment for bovine trichomonosis infection.

Comparison of quantitative polymerase chain reaction (qPCR) test results of cultured smegma samples from feeder and study laboratories and pooled sample testing (1 positive plus 4 negatives) with *Trichomonas foetus* DNA nested PCR and sequencing results.*

Laboratory	True Pos	True Neg	False Pos	False Neg	Total	Statistical analysis						
						Se	Sp	PVP	PVN	Observed agreement	Cohen kappa (95% CI)	P value†
A	70	295	3‡	6	374	0.921	0.990	0.959	0.980	0.975	0.92 (0.88–0.97)	<0.0001
B	21	62	7	10	100	0.677	0.899	0.750	0.861	0.830	0.59 (0.42–0.77)	<0.0001
C	11§	50	0	2	63	0.846	1.000	1.000	0.962	0.968	0.90 (0.76–1.04)	<0.0001
D	17	29	0	4	50	0.810	1.000	1.000	0.879	0.920	0.83 (0.67–0.99)	<0.0001
F	34	182	0	0	216	1.000	1.000	1.000	1.000	1.000	1.00 (1.00–1.00)	<0.0001
Combined feeder labs	153§	618	10	22	803	0.874	0.984	0.939	0.966	0.960	0.88 (0.84–0.92)	<0.0001
Heat-lysis extract	108	386	10	20	524	0.844	0.975	0.915	0.951	0.942	0.84 (0.79–0.90)	<0.0001
Chemical extract	45	232	0	2	279	0.957	1.000	1.000	0.991	0.992	0.97 (0.94–1.00)	<0.0001
Study laboratory	175	625	3‡	0	803	1.000	0.995	0.983	1.000	0.996	0.996 (0.98–1.00)	<0.0001
1/5 pools	169	50	0	7	226	0.960	1.000	1.000	0.877	0.969	0.91 (0.85–0.98)	<0.0001

* Pos = positive; Neg = negative; Se = sensitivity; Sp = specificity; PVP = predictive value of positive test; PNP = predictive value of negative test; CI = confidence interval; extract = extraction method. Nested PCR and sequencing were carried out on all 178 positive qPCR samples identified in the study laboratory and on 56 negative qPCR samples (n = 234).
 † The P values reported in the table are based on the null hypothesis that the agreement between the feeder and study laboratories in the population is purely due to chance (H₀: kappa = 0).
 ‡ One sample was classified as positive in both the study laboratory and the feeder laboratory but was unable to be sequenced. This sample was considered a false-positive test result in both laboratory A and the study laboratory for this analysis.
 § Inconclusive and presumptive positive classifications by the various laboratories were classified as positive tests in this analysis.

Ordering information

Product	Type	Quantity	Cat. No.
VetMAX-Gold Trich Detection Kit	Real-time PCR	100 reactions	4483869
Workflow products			
VetMAX-Plus qPCR Master Mix	Sample prep	100 reactions	4415327
KingFisher Flex Magnetic Particle Processor with 96 Deep-Well Head	Sample prep	1 instrument	5400630
Applied Biosystems 7500 Fast Real-Time PCR System with Dell Notebook	Analysis	1 instrument	4365464
Related products			
VetMAX-Gold BVDV PI Detection Kit	Real-time PCR	100 reactions	4413938
VetMAX MAP (Johne's) Reagents	Real-time PCR	100 reactions	4405545
VetMAX MAP (Johne's) Controls	Real-time PCR	Varies	4405546

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