

Notes on Quality

Too much fecal material can ruin the test by making the medium too cloudy for examination. When necessary, subculture the suspect InPouch tests into another InPouch.

While differential staining can sometimes be of help in *Tritrichomonas* species identification based on the number of flagella, PCR testing is the only reliable means of definitive identification.

In bovine samples *P. hominis* or other non *T. foetus* protozoa are contaminants.

Reading the Results

Evaluation

To search for the presence of microorganisms, place a viewing clip horizontally over the lower chamber of the InPouch and close (the clip is optional). Place InPouch on the microscope stage under low power (100x magnification) to look for trichomonads. Use a higher power (200x-400x magnification) if necessary for confirmation.

Observation of 1 to 10 live, motile *T. foetus* is all that is required for a presumptive positive result. Continue incubation and repeat the microscopic observation daily for six days before a negative result is reported.

Field studies indicate that 98% of positive results will occur within 5 days; up to 2% of positive results may not be detected until the 6th day of incubation/observation.

Reading Tips

For immediate wet mount examination: Before expressing sample to lower chamber and prior to incubation, roll the top edge down twice and fold over the end tabs to seal the pouch; observe the sample in the top chamber microscopically.

- *Tritrichomonas* gravitate to the bottom and side edges of the pouch chamber.
- Verify that your field of focus is in the liquid and not the textured plastic film layer of the pouch.
- Do not mistake Brownian motion or small particles for evidence of *Tritrichomonas* activity. *T. foetus* are relatively large (9-20 µm) and highly motile.

Additional Product Notes

- NEVER refrigerate or freeze the specimen when culturing. Refer to reference lab sample acceptance requirements if results are to be confirmed by PCR testing.
- Complete each label with the sample information.
- All specimens should be handled according to CDC-NIH recommendations for Biosafety Level 2 (BSL-2) organisms.

Scan for additional product information



References

1. BonDurant. Vet Clin North Am Food Anim Pract. 1997. 13(2):345-61
2. Thomas, et al. Agri-Practice. 1990. 11:13-17
3. Borchardt, et al. Veterinary Medicine. 1992. 11:104-112
4. OIE – World Organisation for Animal Health. OIE Terrestrial Manual. 2018. 3.4.15: 1210-1221

Tritrichomonas foetus LIVE CULTURE

Live cultures of *T. foetus* (clinical isolate) for research, training and QC purposes are available (N. American customers only). This live culture (positive control) can be purchased from Biomed Diagnostics (Catalog No. 12-021-003).

To maintain an active culture, inoculate a new pouch with one drop (approximately 40 µl) of the actively growing culture and incubate at 37°C for 24 hours. They can then be moved to a 32°C incubator or to 18–25°C. Subculture every 3-4 days when the organisms reach a concentration of 1×10^5 /mL.

Symbol glossary: biomeddiagnostics.com/l/symbol-glossary

Document Revision History

Rev. J, August 2019

New format; added new catalog numbers, °F, document revision history; removed "For In Vitro Diagnostic Use Only"; specified 18–25°C instead of room temperature; reorganized and retitled some sections

Rev. K, October 2019

Removed °F; corrected typos and terminology; consolidated "Materials Required but Not Provided"; added OIE reference.

Manufactured by:
Biomed Diagnostics, Inc.
1388 Antelope Road
White City, OR 97503 USA
biomeddiagnostics.com



InPouch® TF - Bovine

A selective culture system for the diagnosis of
Bovine *Tritrichomonas foetus*

| | | |
|-------------|------------|-----------|
| Catalog No. | 12-011-001 | 10 Tests |
| Catalog No. | 12-011-002 | 100 Tests |

For Veterinary Use Only

Download



Certificate of Analysis

Introduction

Intended Use

InPouch® TF is a self-contained system for the detection of *Tritrichomonas foetus* from bovine preputial or vaginal samples. The proprietary medium is selective for the transport and growth of *T. foetus*, while inhibiting the growth of other organisms that can interfere with a reliable diagnosis.

Description and Principle

Bovine trichomonosis is a venereal disease transmitted by the protozoan parasite *T. foetus*. The primary pathological manifestation of this infection is early embryonic death or abortion in impregnated cows. Cows show few other symptoms of infection, while bulls are asymptomatic.

InPouch TF is designed to facilitate and simplify the detection of *T. foetus* as the organism is infrequently found in direct microscopic examination of clinical specimens, and serological methods of diagnosis are not reliable. This device conveniently supports the following user needs in a single-exposure system:

- Ease of inoculation
- Proprietary medium selective for TF growth
- Direct microscopic observation of the culture
- Self-contained culture system
- Incubatory capabilities
- Safe transport and preservation of the specimen
- PCR compatible

Reagents and Appearance

InPouch contains peptones, yeast extract, maltose and other nutrients, amino acids, salts, antifungal and antimicrobial agents in a phosphate buffered saline base constructed to isolate the positive detection of *T. foetus*. Final pH of media is 6.7 ± 0.1 at 25°C .

Specificity

InPouch TF medium is known to be effective in culturing *T. foetus*, *T. suis*, *T. gallinae* and *P. hominis*.

Precautions, Safety and Disposal

Read the Safety Data Sheets (SDSs) and follow the handling instructions. Wear appropriate protective eyewear, clothing and gloves.

InPouch TF is for veterinary protozoa identification and test results only.

Consult your local (e.g., State Department of Agriculture) regulations before use. Some states require that only certified veterinarians collect and read bovine TF cultures and/or submit samples for PCR testing.

InPouch's growth medium suppresses but does not eliminate yeast and bacterial growth. A build-up of gas from bacterial growth can be vented by opening the pouches inside a BSL-2 rated biological safety cabinet.

WARNING: This product contains chemicals known to the State of California to cause cancer, birth defects and other reproductive harm.

Once the pouch has been inoculated and resealed, re-open only in a biological safety cabinet. Because of the potential for containing infectious materials, the pouch must be destroyed by autoclaving at 121°C for 20 minutes or other suitable means for sterilization and disposal of BSL-2 organisms.

Procedure

Key Notes Regarding Specimen Collection

Sample Preparation

Clip the hair around the preputial orifice in bulls. Flush the preputial cavity with sterile saline solution (not water) to clean out mud and manure if necessary (decreases the risk of overgrowth of non *T. foetus* bacterium).

Collect Sample

Bulls: Direct the pipette to the distal penis in the sheath. Scrape the mucosa of the distal penis of the fornix area while applying suction with the syringe or bulb to obtain the specimen.

Cows: Advance the pipette gently to the floor of the vaginal fornix and aspirate mucus.

Materials Provided

- InPouch TF test(s)

Materials Required but Not Provided

- Sample (see "Key Notes Regarding Specimen Collection", above)
- Infusion/insemination pipette
- 20 mL syringe/pipette bulb per bull/cow
- Laboratory incubator capable of incubation at $30\text{--}37^{\circ}\text{C}$
- Microscope ($\geq 100\times$)

Storage

Do not refrigerate or freeze InPouch TF tests. Upon receipt, store at $18\text{--}25^{\circ}\text{C}$ horizontally, away from direct sunlight. Do not use expired tests. Do not use an InPouch test if the liquid appears to be cloudy, leaky, dark brown or dried.

Shelf Life

InPouch TF - Bovine has a shelf life of 12 months from the date of manufacture.

Incubation

Incubate the pouch vertically at $35\text{--}37^{\circ}\text{C}$ for up to 6 days. InPouch TF is designed for safe transport, if needed. Inoculated InPouch TF tests should be transported within 48 hours after inoculation and maintained at $15\text{--}37^{\circ}\text{C}$.

Quality Control

This product has been tested and meets the CLSI (formerly NCCLS) Approved Standard for commercially prepared media (M22-A3). At the time of manufacture, quality control testing is performed on each

Prepare InPouch



Remove the pouch from the bag and manually express the liquid so that all the liquid is in the lower chamber. Open the pouch by tearing off the top. There is a pre-formed score to facilitate tearing. Use the integral white tabs to open and secure the mouth of the pouch open.

Inoculate Sample



Insert the sample into the upper chamber of the pouch (0.5-1.0 mL of specimen). Squeeze a small amount of liquid from the lower chamber to the upper pouch chamber to flush the sample. Minimize the introduction of bubbles or foam. Dispose of pipette and syringe.

Integrate Sample



Express the entire content of the InPouch into the lower chamber. Avoid trapping air. Roll the pouch top tightly, until the wire-tape is at the top of the label. Fold the wire-tape end tabs to seal the pouch.

lot of InPouch TF. The ability of the media to support expected growth, selectivity and morphology is verified by lot.

InPouch TF is manufactured in accordance with controlled procedures at Biomed Diagnostics. Each lot undergoes an initial QC performance testing prior to release for consumer use. Additional performance testing is repeated at specific intervals throughout the marked shelf-life of each lot to ensure absolute reliability of the product.

The following is recommended for customers who choose to complete independent QC testing of InPouch TF:

- Obtain a sample of viable *Tritrichomonas* organisms in the range $2.0 \times 10^5\text{--}2.0 \times 10^6$ live cells/mL.
- Inoculate three (3) InPouch TF diagnostic tests with 1-2 drops of the live culture using a sterile glass Pasteur pipette (~20–40 μl) per the "Inoculate Sample" steps discussed in this insert.
- Incubate the inoculated InPouch TF diagnostic tests for 24 hours at 37°C .