



Quality Control Procedure for the InPouch™ TF Diagnostic Test

The InPouch™ TF diagnostic test is manufactured in small lots. BioMed Diagnostics guarantees performance of all its diagnostic tests up to the printed expiration date on each test device. Each lot is released by Quality Assurance (QA) only after in-house verification of sensitivity, specificity and sterility. Additionally, QA continues this testing throughout the product's labeled shelf-life, re-testing each lot at 3-month intervals until the expiration date. BioMed Diagnostics Inc. utilizes its own rigorous testing methods, which are based, in part, on CLSI (M22-A3) Standards and Recommendations and other pertinent regulations. For further details contact BioMed Diagnostics.

In addition to ATCC reference strains, BioMed Diagnostics maintains live clinical-isolate reference cultures of *T. foetus*, which are used for QA testing of the InPouch™ TF diagnostic test. Live cultures are maintained in our QA laboratory. Live *T. foetus* cultures are available for purchase through BioMed Diagnostics (transport of live cultures is unavailable in some countries).

We recognize that customers may, from time to time wish to perform their own quality control testing to verify performance, to satisfy regulatory requirements and for general trouble shooting. This document shall serve as an official quality control guide for customers and end-users.

Safety

According to CDC guidelines, *Trichomonas* species. are Biosafety Level 2 (BSL-2) infectious organisms. Open live cultures only in a BSL-2 hood, dispose of old culture pouches and inoculation material in a biohazard container, disinfect all surfaces and clean up any spills according to BSL 2 procedures, and/or follow your laboratory's BSL-2 procedures for handling and disposing of these organisms. Only trained, qualified personnel should perform this procedure. Please review the InPouch™ TF product insert and instructions before proceeding. See: <http://www.cdc.gov/OD/ohs/biosfty/bmbl4/bmbl4s3.htm> for more information about Biosafety Level Criteria.

Equipment and Materials

- Lab attire: lab coat, protective gloves, safety glasses
- Biohazard container
- BSL-2 rated workspace
- 37° C incubator
- Microscope with 10x objective (100x magnification)
- Hemocytometer
- Glass pipettes - sterile
- InPouch™ TF diagnostic tests (3 each from each lot to be tested)
- Live TF culture (source pouch), 1-2 day culture grown at 37° C

Procedure

- Remove the live culture source from the incubator and thoroughly suspend the organisms. If using a live culture from BioMed Diagnostics, agitate the medium by lightly pulling the InPouch™ up and down across the edge of a table or counter top in a 'shoe shine motion' 5 times. Avoid creating bubbles.



- Examine the live culture microscopically (10x objective). Use a hemocytometer to confirm that you have viable *Trichomonas* organisms in the range of 2.0×10^5 - 2.0×10^6 live trichomonads/mL. If there are too few, incubate the source pouch an additional 12-24 hours. Record the results for future reference.
- Take 3 preselected InPouch™ TF diagnostic tests from the lot to be tested. Label them A-C; Be sure to include inoculation date as well as the strain of *T. foetus* used. Perform a visual inspection of the InPouch™ TF diagnostic tests before inoculation; In case of cloudiness, low volume, or discoloration, do not use, and contact BioMed Diagnostics immediately.
- Open the InPouch™ TF diagnostic tests by tearing the top off of the InPouch™ and pulling the two tabs apart.
- Inoculate each InPouch™ TF diagnostic test with 1-2 drops of the live culture using a sterile glass Pasteur pipette (~20-40 µl).
- Fold the opening of the InPouch™ TF diagnostic test over one time and mix the 1-2 drops with the rest of the media by performing the 'shoe shine motion' to ensure proper dilution of the inoculum.
- Close the InPouch™ TF diagnostic tests according to the package insert.
- Incubate all InPouch™ TF diagnostic tests for 24 hours at 37°C.
- After 24 h, remove all InPouch™ TF diagnostic tests from the incubator and resuspend by pulling the InPouch™ up and down across the edge of a table as described in the insert.

Acceptance criteria: Examine each InPouch™ TF diagnostic test microscopically (10x objective). Use a hemocytometer to confirm that you have viable *Trichomonas* organisms in the range of 2.0×10^3 - 2.0×10^6 live trichomonads/mL.

Note: When working with the InPouch™ TF diagnostic tests from the incubator, accurate results may be compromised if the live cultures are left at room temperature longer than 10-15 minutes. The activity of the organisms slows at cooler temperatures, making it difficult to visually identify viable organisms.

- In the case that the 24 h density falls below 2.0×10^3 live trichomonads/mL, incubate the InPouch™ TF diagnostic tests an *additional* 24 h. Examine each InPouch™ TF diagnostic test microscopically (10x objective). Use a hemocytometer to confirm that you have viable *Trichomonas* organisms in the range of 2.0×10^3 - 2.0×10^6 live trichomonads/mL.

Worksheet

InPouch™ TF diagnostic test lot # _____

Sample	Date/Time	Hemocytometer Count	Dilution factor	24 h Density (trichomonads/mL)	48 h Density (trichomonads/mL)
Live culture (source pouch)					
A					
B					
C					