$\mathcal{M}^{\scriptscriptstyle{\mathsf{M}}}$

ST112R. Troph-o-Glo™, FL, 1x conc., Reagent Only

FL-labeled Antibody Reagent for Direct Immunofluorescence Detection of Giardia trophozoites in Stool Specimens

Explanation: Giardia lamblia is a common, ubiquitous intestinal parasitic protozoan that causes gastroenteritis in man and lower animals. This organism has a reservoir of host animals and can be spread through food, water, and fomites. The trophozoite stage of *Giardia* is a teardrop-shaped organism measuring approximately 8-16 um in length and approximately 5-12 um in width, anteriorly. Generally, trophozoites will not be found in environmental samples, however, they may present in fecal specimens.

Description of Products

- This kit utilizes the principle of direct immunofluorescence. The antibody reagent consists of a fluorescein (FL)-labeled monoclonal antibody made to outer trophozoite antigenic sites (epitopes) of Giardia lamblia. The reagent will bind to trophozoites of Giardia lamblia if they are present. The trophozoites will appear bright apple-green when viewed under a fluorescence microscope using a fluorescein filter set.
- The reagent consists of 3.5 mL of a working-dilution (1X) solution of fluorescein (FL)-labeled monoclonal IgG antibody prepared against trophozoites of *Giardia lamblia* derived from cell culture. The volume provided is enough reagent for 75+ tests using one drop per test on well slides, approximately 45 uL per drop. The antibody reagent contains 0.04% w/v sodium azide as preservative and 1% bovine serum albumen as antibody stabilizer.
- > No positive control is provided in this kit. The recommended that the positive control is prepared using freshly cultivated trophozoites.

Storage: Store at 4° C. DO NOT FREEZE. ST112R reagent is light sensitive.

Kit Includes

• ST112R: 1 dropper vial containing 3.5 mL working dilution (1x) reagent

Other Lab Supplies Not Included, but Available

- B100-40: 40 mL Dilution/Blocking (D/B) Buffer
- C101: 3.5 mL BlockOut™ counterstain
- D101: 0.4 mL DAPI, 5000X in methanol
- M101: 3.5 mL No-Fade[™] Mounting Medium
- M102: 3.5 mL Elvanol No-Fade™ Mounting Medium
- S100-1-9MM: One-well (9mm) SuperStick[™] Slides, 40/box
- S100-1: One-well (14mm) SuperStick[™] Slides, 40/box
- S100-2: Two-well SuperStick[™] Slides, 40/box
- S100-3: Three-well SuperStick[™] Slides, 40/box
- WB100: 50 mL 20x SureRinse[™] Wash Buffer
- WB101: 50 mL 1x SureRinse™ Wash Buffer
- PACIR: AccuSpike™-IR, G/C Quality Control Standard (PACIR3, PACIR6, PACIR12)

Preparation

1. Prepare environmental sample(s) to be applied to well slide.

Contact us by email for MSDS or Certificate of Analysis/QC Report. Email: contact@waterborneinc.com

Instructions for Use

- 1. Isolated water particulates should be air-dried onto a well of a pre-treated slide, using a stream of warm (not hot) air; alternatively, a slide-warmer may be used. Do not allow the slide to become hot to the touch. Samples must be completely dry before continuing to step 2. (Drying time: Approximately 15 30 minutes.)
- 2. Apply one drop (approximately 45 uL) of Troph-o-Glo™ G/C antibody reagent to the spot of dried test particulates in each well. If necessary, spread the drop with applicator stick or glass rod, being careful not to contact the surface of the slide.
- Incubate the slide in a humid chamber at room temperature for at least 25 minutes. If using a 37° C incubator, incubate for 25 minutes. Longer incubation periods are OK.
- 4. Rinse the slide free of antibody reagent by adding 50 100 uL SureRinse™ wash buffer and leave for 1 minute. Tilt slide, long edge down, and absorb excess fluid with absorbent material placed at the edge of the slide well. Do not touch the surface of the well slide or disturb the sample.
- 5. Non-specific background fluorescence may be reduced, and a reddish background added to enhance contrast, by the use of BlockOut™ counterstain at this stage. Apply 1 drop of counterstain per well. Incubate for 1 minute at room temperature.
- 6. Rinse the slide free of counterstain by adding 50 − 100 uL SureRinse[™] wash buffer and leave for 1 minute. Tilt slide, long edge down, and absorb excess fluid with absorbent material placed at the edge of the slide well. Do not touch the surface of the well slide or disturb the sample.
- The slide should be partially-to-completely air dried on a slant and then mounted with one drop (~45 uL) of No-Fade™ mounting medium. Apply cover glass and view.

Other Information, Tips & Troubleshooting

- Test Time: Approximately 35 40 minutes after the sample is dried to the well slide.
- Prepared slides (mounted with M101, No-Fade™ mounting medium) may be kept in a refrigerator/protected from light and viewed repeatedly for 6 months or longer. DAPI staining may fade.
- 3. One resource available to help distinguish between *Giardia* cysts, *Cryptosporidium* oocysts and possible cross-reactors can be found on the US EPA website. The US EPA has developed training modules for the Long Term 2 (LT2) Enhanced Surface Water treatment Rule. These training modules were developed to assist analysts in the detection and identification of *Giardia* and *Cryptosporidium*.

They can be found at: www.epa.gov/safewater/lt2/training/index.html.

For assistance, technical questions, or to inquire about other Waterborne $^{\mathsf{TM}}$, Inc. products, please call, FAX, or e-mail us. Also, please visit our website at www.waterborneinc.com.

Reference(s):

¹ Bogitsh, B. and Thomas Cheng. <u>Human Parasitology</u>. Saunders College Publishing, Philadelphia. 1990.