



## A100FLK. Aqua-Glo™ G/C Direct Comprehensive Kit

Fluorescein-labeled Monoclonal Antibody Reagent for Simultaneous Direct Immunofluorescence Detection  
Of Giardia Cysts and Cryptosporidium Oocysts in Water Samples  
*Aqua-Glo™ G/C is EPA - approved for use in Methods 1622 and 1623.*

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**Explanation:** Giardia lamblia and Cryptosporidium parvum are common, ubiquitous intestinal parasitic protozoa that cause gastroenteritis in man and lower animals. Both organisms have a reservoir of host animals and can be spread through fecal contamination of food, water, and fomites. The Cryptosporidium oocyst is a nearly round encysted organism of approximately 3-5 um in diameter, while the Giardia cyst is oval-shaped and measures approximately 8-13 um in length and 7-10 um in width.

**Biological Principles & Description of Products:** The Aqua-Glo™ kit is designed to detect the cyst and oocyst stages of these parasites in particulates isolated from water and other environmental samples utilizing the principle of direct immunofluorescence. The **antibody reagent** included consists of a mixture of fluorescein-labeled mouse monoclonal antibodies made to cyst and oocyst outer wall antigenic sites (epitopes) of Giardia lamblia and Cryptosporidium parvum. The reagent shows varying degrees of cross-reactivity with cysts and oocysts of other species of Giardia and Cryptosporidium. The cysts and/or oocysts will appear bright apple-green when viewed under a fluorescence microscope using the appropriate filters for fluorescein. The reagent is genus-specific and will bind only to the cysts and oocysts of these two parasites if they are present. **Counterstain** contains Evan's Blue with 0.04% w/v sodium azide as preservative. **Mounting Medium** is fade retardant. **Positive Control** is a mixture of Giardia lamblia cysts and Cryptosporidium parvum oocysts in 1.0% formalin in PBS. The concentration of this suspension is approximately 2x10<sup>5</sup> cysts and oocysts (each) per mL. 20x wash buffer is a saline solution provided for the rinsing process. SuperStick™ Slides are chemically treated to increase cell adhesion. The wells measure 15 mm in diameter.

### Kit Includes:

- 1 dropper vial containing 3.5 mL working dilution (1x) reagent
- 1 dropper vial containing 3.5 mL counterstain (catalog # C101)
- 1 dropper vial containing 3.5 mL No-Fade™ Mounting Medium (catalog # M101)
- 1 glass vial containing 1 mL positive control
- 1 screw cap bottle containing 50 mL 20x wash buffer
- 1 box of two-well SuperStick™ Slides, 40/box (catalog # S100-2)

### Other Lab Supplies Not Included, but Available:

- S100-1: One-well SuperStick™ Slides, 40/box
- S100-3: Three-well SuperStick™ Slides, 40/box
- M102: 3.5 mL Elvanol No-Fade™ Mounting Medium

**Storage:** Store at 4° C. DO NOT FREEZE!

### Instructions for Use:

1. Isolated water particulates should be air-dried onto a well of a slide, approved for use in the method, 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.
2. When the sample has dried, a methanol fixation step may be performed at this point, as recommended in EPA Method 1623; however, methanol fixation is not required for this reagent to bind well to cysts and oocysts.
3. When the sample has dried **completely**, apply one drop (approximately 45 uL) of **Aqua-Glo™ G/C** antibody reagent to the 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.
4. Incubate the slide in a humid chamber at room temperature for at least **40 minutes**. If using a 37 deg C incubator, incubate for 30 minutes. Longer incubation periods are OK.
5. Rinse the slide free of antibody reagent by soaking the slide for at least **2 minutes** in 1x saline wash solution or in "PBS" (phosphate-buffered saline).
6. Non-specific background fluorescence may be reduced, and a reddish background fluorescence added to enhance contrast, by the use of counterstain at this stage. Apply 1 drop of counterstain per well. (Incubate for one minute. Follow with a 1-minute rinse in PBS or saline. Longer incubations are OK.)
7. Following a rinse, the slide should be partially to completely air-dried on a slant and then mounted with appropriate mounting medium containing an anti-fading agent. Complete mounting with a cover glass and view.

For assistance, technical questions, or to inquire about other Waterborne™, Inc. products, please call, FAX, or e-mail us. Also, please visit our website at [www.waterborneinc.com](http://www.waterborneinc.com).

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